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TRAGOPOGONS AS WEEDS IN CANADA[1]

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The Tragopogons (Family Compositae) are Old World plants ranging from the western Mediterranean region to western Asia and Siberia, several species also having become established in North America. These latter are all stout, erect, smooth plants, commonly from one to three feet in height, with flower heads and fruit terminating the branches and bearing some resemblance to those of dandelion. Stalks are produced from more or less fleshy, biennial tap-roots.

HISTORY

The earliest available Canadian reference is one to *Tragopogon porrifolius* L. by Provancher (10) and this only to its cultivation as salsify or vegetable oyster. As an escape *T. pratensis* L., the yellow goatsbeard, was recorded in 1867 by Hubbert (7). Billings' list for Ottawa in 1866 (2) did not include either plant but Fletcher (4) refers to both, the specimens having been collected respectively by himself and his associate, J. A. Guignard in 1879, and remaining in the herbarium of the Division of Botany, Ottawa. The original station for *T. pratensis* was "Along the railway track near the St. Louis dam, Ottawa." Older residents will recall this as the north end of Dow's Lake, now a portion of the Federal District Commission driveway. Occurrence along this section of railway is still about as abundant as anywhere in the city and district.

Macoun (9) assembled records also for *T. pratensis* at St. Stephen, N.B. and Pictou, N.S.; and for *T. porrifolius* at Belleville, London and Strathroy, Ont. In Part III of his Catalogue (1886) he added for *T. porrifolius:* "Around Lotbiniere, Que. (*St. Cyr*). Victoria, Vancouver Island. (*Fletcher*)." Both plants have become more generally distributed but remain comparatively unknown to the general public.

Rydberg in 1917 (11) listed a third species, *T. dubius* Scop. (known as greater goatsbeard in Europe), as being escaped in Colorado. A note by Hull (8) recording its detection in Indiana in 1943 carries an editorial footnote: "This large-headed species, *Tragopogon dubius* Scop. (1772), including *T. major* Jacq. (1773) has recently been sent to the Gray Herbarium from Virginia, Michigan, Illinois, Minnesota, South Dakota, Oklahoma, Texas, Washington, Oregon and California." *T. major* here, and as used by some American botanists, appears to be equivalent to *T. dubius* Scop. as treated by Hegi (6) to equal *T. major* Jacq. and *T. dubius* Scop. subsp. *major* (Jacq.) Vollm. If two entities are present here they have not been clearly separated.

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DISTRIBUTION

Old World distribution of the three plants is not closely conterminous. *T. porrifolius*, being widely cultivated, is also in such places escaped. *T. pratensis* ranges, according to Hegi (6), from Ireland, Denmark and much of Scandinavia to northern Karelia in Russia, on the north, to middle Spain, Italy, Macedonia and south Russia; also in the Caucasus, Armenia, western Persia and Siberia. *T. dubius* is in northern France, across Germany to West Prussia and Poland, and southward to Spain, middle Italy, the Balkans and south Russia, and extending to Asia Minor and the Caucasus; a generally less northern range than that of *T. pratensis* although in Valais, southern Switzerland, where both occur, reaching a little higher altitude than the other.

In North America about the same situation holds with regard to latitude. *T. pratensis* ranges, according to the manuals, from New Brunswick to Manitoba and south to New Jersey and Colorado. *T. dubius*, as as well as in the States enumerated above, extends northwestward into Canada, a zonal trend common among biological forms and not inconsistent with the conclusion that *T. dubius* occupies the less high latitude of the two.

In Canada *T. dubius* was being confused at first with *T. pratensis* but has been distinguished in the western provinces for about ten years. A specimen was collected by the writer at Beaverlodge, Alta., in 1934; and another, with immature flower, secured at Lethbridge, Alta., in 1929 appears to be this. In the East it was not detected until 1943, although a specimen sent from Rondeau Park in 1942 and another collected near Brantford, Ont., a year earlier by W. H. Minshall both prove to be this species. In the Ottawa district suspected presence of both yellow-flowered goatsbeards was confirmed when they were found in 1945 growing together at Westboro between the C.P.R. tracks and the Ottawa River, and for two miles into the city. Both species were also growing two miles south near City View and across the river at the Hull Armouries and near the Canada Cement Company quarries, a total span, north and south of about five miles. Scouting up to six or eight miles in all directions from the city has failed to locate any more *T. dubius* although the longer known plant is found along most roads. About the same date a second Quebec station was found for *T. dubius* by C. Frankton near Ville St. Pierre, outside of Montreal. Later in the summer a specimen from Fitzroy Harbour, Ont., was found to be this species; and a light field infestation five miles east of Renfrew still further extended the known range in eastern Ontario. With closer discrimination than formerly, specimens have now been added to the Division herbarium from seven counties in Ontario: Renfrew, Carleton, Hastings, York, Simcoe, Brant and Kent; and in Quebec from Gatineau and Jacques Cartier counties.

From herbarium evidence *T. pratensis* would be in Ontario the predominant, and eastward practically the only, species. On the prairies, however, and in the drier parts of British Columbia *T. dubius* is the more prevalent. On a survey west to Saskatoon, Rosetown and Regina in early summer, 1945, it was first recognized near the Ontario-Manitoba boundary and was the only one seen westward. According to Russell (5) it "has become much more abundant and widely distributed in the province (Saskatchewan) during the past ten years."

INCIDENCE

Both species tend to be in colonies in this country as well as in Europe. *T. pratensis* is reported to occur in its native habitat from somewhat moist, well-fertilized meadows to drier slopes, waysides, waste places and occasionally in fields; *T. dubius* on dry stony slopes and even quarries, walls and road embankments, and freight yards, as well as in open woods, stream banks, meadows and vineyards. This species, perhaps both, are commonly on chalk or lime. The impression conveyed is that *T. dubius* endures generally drier, more adverse conditions than the other species, and this, broadly, is what is indicated in Canada.

Weed surveys, while well able to distinguish Tragopogons from other plants and, at least when in flower, to distinguish *T. porrifolius* from the others by colour, were not, unfortunately, in earlier years taking account of two yellow goatsbeards. For this reason survey data for these two are merged in Table 1. By meridional belts percentage incidence (surveys of each 100 in which recorded) is seen to vary from almost nil in the central belts to figures for the rest still low as compared with most noxious weeds.

TABLE 1.—PERCENTAGE INCIDENCE IN CANADA

131-116° B.C. and Peace R. 642 surv.	115-108° Alta. and W. Sask. 317 surv.	107-100° Sask. and W. Man. 341 surv.	99-92° Man. and W. Ont. 167 surv.	91-84° Superior region 101 surv.	83-76° Cent. Ont. and N. Que. 902 surv.	75-68° E. Ont. and Cent. Que. 840 surv.	67-60° Maritimes and Gaspe 1005 surv.
%	%	%	%	%	%	%	%
T. dubius (west largely) and *T. pratensis*							
9.0	3.7	4.7	2.9	1.0	9.5	11.6	10.5
T. porrifolius							
1.4	.3	0	2.4	0	3.2	0	0

Although 10% may seem light incidence it must be remembered that it consists commonly of rather dense occurrence within more localized distribution than that of many other weeds. All three frequent waysides (road and railway) with increased density in proximity to towns and heavier traffic. Less frequently infestations occur in thin sod and even in crops as shown in Figure 1 where a field was completely over-run by *T. dubius*.

For *T. porrifolius*, a more domesticated plant than the others, lighter incidence is shown, and was found mostly in Ontario, south central Manitoba and in the upper Okanagan and coastal parts of British Columbia. Higher incidence, for *T. pratensis* in the East and for *T. dubius* over much of the West, indicates for these somewhat readier naturalization and, apparently, some ecological specialization although they grow together quite well in Ontario. The failure of *T. pratensis* to colonize the prairies, as *T. dubius* has done in a much shorter period and is doing both eastward and westward, points to the latter as the greater potential weed.

Under Ottawa conditions *T. porrifolius* was seen sparingly only along roadsides through fertile land. The others were in less restricted habitat, and without much dwarfing of either on the shallowest soil or on railway ballast. Almost bare limestone supports them. On the hot, dry plains of the West, plants of *T. dubius* are often forced into bloom at a few inches in height. *T. pratensis*, on the other hand has been seen up to five or six feet in height near Ottawa under the dense shade of roadside trees. Marked sensitivity to sunlight is shown by the bending of heads to the morning sun and their closing by noon unless delayed by a change to dull or rainy weather. Noon-flower or go-to-bed-at-noon are colloquial names expressive of this trait. Commonly the more slender peduncles of *T. pratensis* are found bent and twisted permanently as a result of such daily light response.

DESCRIPTION

Descriptive remarks for those unfamiliar with these species will be confined to the field characters useful for their separation since they are not often confused with other plants. Those found most helpful are summarized in Table 2.

TABLE 2.—FIELD CHARACTERS USEFUL IN IDENTIFICATION OFTEN LOST IN DRIED SPECIMENS

Characters	*T. pratensis*	*T. dubius*	*T. porrifolius*
Flower colour (yellows of Horticultural Colour Chart (3))	Canary yellow	Sulphur yellow	Purple
Peduncle	Little thickened, often flexuous	Thick, stiff, fistulose	Thick, stiff, fistulose
Involucral bracts	Equalling florets, broad, tinged with purple	Longer than florets, narrow, without purple	Longer than florets
Fruit (including beak and, with pappus, determining size of globes)	Mostly under 20 mm. long	Mostly over 20 mm. long	
Stalk	Tending to purplish in full light, especially on nodes	Seldom purplish except at base	
Leaf tips (at all ages)	Usually curved or curled	Straight and grass-like	Straight

WEED CHARACTER

The three species are all offenders against private and civic pride in well-kept surroundings; and *T. dubius* has provided enough instances of field infestation to warrant close vigilance concerning it. They are all prolific of seed which has been found, in the case of *T. pratensis* (1), to have almost perfect germination at maturity and continuing high for several years. Germination in nature seems to be restricted, by seed-bed

requirements no doubt, to just those scarred embankments and waste places, not too densely turfed, which are commonly found infested. Tilled land would provide the seed-bed but, unless sowed to a crop like fall wheat, rye or hay, that leaves it undisturbed through a second season, it does not survive to reproduce. Maintenance of roadsides in close sod and better clean-up of breeding places of weeds would restrict its area. Mowing to prevent seeding should commence with early blooming and needs to be repeated later for secondary growth. Up-rooting is therefore even better and, with such erect plants, is feasible by hand if not by implements.

FIGURE 1. *T. dubius* infestation in crop, upper grass zone, Brigade Lake district near Kamloops, B.C.

SUMMARY

The history, present distribution and status in Canada of three species of *Tragopogon* are discussed. *T. dubius*, of fairly recent introduction and still largely of the drier West, appears to exhibit marked weed tendencies, *T. pratensis* remains still, after three-quarters of a century, mostly a wayside and waste place encumbrance, and *T. porrifolius* a sparing escape from cultivation.

Practical field characters for distinguishing the species are given. Ecology, life history and control are only lightly touched upon pending further investigation. As biennials, with abundant seed of high germination capacity, the infrequence of field infestation is believed due to less success in finding a seed-bed than in loose, often bare, vacant lot and wayside sites.

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CONTRIBUTIONS TO THE STUDY OF RANCIDITY IN CANADIAN CHEDDAR CHEESE[1]

III. A COMPARISON OF THE LIPOLYTIC ACTIVITY OF SLIGHT RANCID AND FIRST GRADE SAMPLES

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In seeking to assess the possible role of milk lipase as a causal agent in the production of rancid cheese, the suggestion was made by Hammer (4) that rancid cheese might show greater fat hydrolysing activity when introduced into sucrose-cream mixtures than would cheese of normal flavour. Investigators of lipase activity in cheddar cheese have so far not studied the problem from the point of view of enzyme activity of the cheese itself (5, 10, 13). Gould (3) has shown that the lipase content of milk is identified with the skimmilk fraction and accordingly one might expect much of this enzyme to pass into the whey. On the other hand, the fact that a slow fat hydrolysis can be demonstrated in raw milk cheese (7, 10) would appear to be sufficient grounds for investigating rancidity in cheese with this point in view.

EXPERIMENTAL

Determinations of lipase activity were made on two sets of samples. The first of these comprised 19 samples, 4 of which were graded 41-94 or higher, while the remainder were all of second grade quality having been scored down as a result of either slight rancid, fruity, not clean, or off flavours. When held for 5 days in a sucrose-cream preparation at 98° F. it was not possible to show significant differences in the acidities of these mixtures. This work was of a preliminary nature and will not be further described.

The second series of samples consisted of three 1-pound blocks of first grade cheese and three 1-pound blocks of second grade, slight rancid cheese. All of these samples were manufactured during the period of October 13-19, 1942 and the analysis was carried out on them between November 13 and December 4. Preliminary to determining lipase activity the samples were analysed for fat by the Mojonnier Method (11), for total solids by the A.O.A.C. method (1), and for pH by means of glass electrode and a Coleman electrcmeter. In addition the acidity of the cheese fat was determined. The determination consisted of grinding about 250 g. of the cheese to be tested in a kitchen food chopper, the ground cheese then being allowed to oil off by placing it in a tall 400 ml. beaker in a water-bath at 125° F. for $2\frac{1}{2}$ to $3\frac{1}{2}$ hours. Twenty grams of the fat obtained by this means was then dissolved in 50 ml. of neutral alcohol, brought to a boil over hot water, and immediately titrated against 0.1N NaOH using 0.5 ml. of phenolphthalein as indicator. This titration has been used by Breazeale and Bird (2) in butter deterioration studies. The results of these determinations are presented in Table 1.

[1] Contribution from the Dairy Department.
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TABLE 1.—VALUES FOR FAT, TOTAL SOLIDS, FAT ACID VALUE AND pH ON SIX SAMPLES OF CHEESE STUDIED FOR LIPASE ACTIVITY

Sample	Flavour	Fat	Total solids	Acidity of cheese fat	pH
		%	%		
1	Clean	34.17	65.95	2.85	5.18
2	Clean	36.02	67.85	2.30	5.17
3	Clean	34.50	65.51	2.53	5.04
4	Sl. rancid	34.81	65.76	3.33	5.11
5	Sl. rancid	35.20	65.84	2.70	5.05
6	Sl. rancid	34.92	66.60	2.75	5.20

The sucrose-cream mixture was prepared as follows. Cream testing 37% fat was secured and the amount of water present calculated. Cane sugar in an amount equal to twice the weight of the water present was then stirred in until dissolved. To facilitate solution of the sugar, the mixture was heated to 150° F. after which it was held for 1 hour. While still at this temperature, the mixture was homogenized by passage through an "Empire" cream maker. The reaction was then brought to pH 8.2 by the addition of 0.2N NaOH solution after which it was dispensed into 6-ounce bottles in 100 ml. amounts. These were then held in an atmosphere of flowing steam for 1 hour. The method is adopted from that of Roahen and Sommer (12).

Two 10-g. portions of each sample of cheese were added to separate 100 ml. quantities of sucrose-cream mixture. To facilitate homogeniety, the cheese and warmed sucrose-cream were ground together in a mortar. Immediately thereafter, one of this pair of bottles was held in a boiling water bath for 15 minutes to destroy any lipase present. Both bottles were then placed in a water bath and held for 12 days at 98° F. All samples were shaken vigorously daily for the first 6 days.

The changes in the acidities of the mixtures were determined by direct titration on a portion of the cheese and substrate as well as by determining the acidity volatile by steam distillation.

The direct titration was carried out by weighing out 20 g. of cheese and substrate, diluting with 20 ml. of distilled water, heating for 1 minute in a boiling water bath and then titrating against 0.1N NaOH solution with 0.5 ml. of 1% phenolphthalein solution.

In the steam distillation, distilled water was used as a source of steam. The apparatus was cleaned by passing steam through the still-head and condenser until 150 ml. of distillate was collected, prior to each determination. Twenty grams of cheese and substrate was adjusted to approximately pH 2 with 10% (by volume) H_2SO_4 solution after which distillation was continued until 100 ml. of distillate had been collected. The distillate was then titrated against 0.1N NaOH with 0.5 ml. of phenolphthalein as indicator. The results are presented in Table 2.

TABLE 2.—PRODUCTION OF ACIDITY IN SUCROSE-CREAM BY NORMAL AND SLIGHT RANCID CHEESE AFTER 12 DAYS INCUBATION AT 37° C.

Sample*	pH after 12 days at 98° F.	pH to which adjusted before distillation	Titration of 100 ml. of distillate	Direct titration of 20 g. of cheese and substrate
1 A	—	—	—	14.03
1 B	—	—	—	6.15
2 A	5.02	1.9	3.07	14.76
2 B	5.7	1.9	1.45	6.15
3 A	5.20	1.75	2.98	12.67
3 B	5.75	1.80	1.50	6.24
4 A	4.95	2.1	2.60	12.26
4 B	5.65	2.0	1.45	5.99
5 A	5.20	2.1	3.03	14.17
5 B	5.82	2.0	1.50	6.42
6 A	5.20	2.0	3.10	13.30
6 B	5.95	1.8	1.50	5.40

* A samples unboiled; B samples boiled. Samples 1, 2 and 3 first grade flavour; samples 4, 5 and 6 slight rancid flavour.

The values presented in Table 2 demonstrate clearly that lipolysis as indicated by the acidities produced in sucrose-cream mixtures was certainly no greater in rancid than in normal flavoured cheese. The results as shown by changes in pH, acids volatile by steam as well as the values obtained by determining the acidity of the cheese and substrate, are all in accord.

DISCUSSION

Two causes have been advanced as possible agents producing rancidity in cheese. The present study has been planned to learn whether normal and slight rancid cheeses would display different lipolytic activities when incubated in a suitable substrate. The results indicate that lipolytic activity is no greater in samples displaying typical slight rancid flavour than in those of normal, first grade flavour.

The treatment which the substrate received (low pressure homogenization) was such as to render it readily attachable by lipase enzyme. Lane and Hammer (9, 10) found that low homogenization pressures were effective in producing rancidity and increased volatile acid production in blue cheese and cheddar cheese. Assuming, therefore, that in this experiment the butterfat was as readily hydrolysed by the lipase in the normal as in the slight rancid cheese, the conclusion is apparent that the two types of cheese contained relatively the same quantity of lipolytic agent.

Krukovsky and Herrington (8) have advanced an "activation" theory to explain the increased rate of lipase action which is brought about in milk by warming and cooling. Hlynka, Hood and Gibson (5) have found that agitation of milk at about 86° F. accelerates the development of rancidity in cheddar cheese. The latter group have also shown (6) that

this agitation results in a greater degree of fat dispersion in the milk. Their contention that the term "activation" should apply more particularly to the substrate than to the enzyme would seem to be valid.

The results reported in the present study appear to suggest that the treatment which the substrate (i.e. the fat globules) receives is the determining factor in the development of this flavour defect, rather than the concentration of lipase. Any and all factors which operate to de-surface the fat globules or increase their state of dispersion, will require study if a solution of this problem is to be achieved.

SUMMARY

Lipase action has been suspected as an agency causing cheese to develop slight rancid and rancid flavours. An experiment is reported in which samples of normal and defective cheeses were incubated in a sucrose-cream substrate. The results indicate that the concentration of lipase was approximately equal in these two types of cheese.

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EFFECT OF CERTAIN METHODS OF HANDLING UPON THE BACTERIAL CONTENT OF DIRTY EGGS[1]

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During the summer of 1944, and again to a lesser extent during 1945, an occasional egg broken in plants breaking eggs for the Special Products Board for subsequent drying was found to contain enormous numbers of bacteria while showing no evidence of abnormality in appearance or odour. The organisms isolated were common soil and water types which presumably had gained entrance to the egg subsequent to its being laid (8).

While the studies of Haines (3) indicate that a good quality, fresh, unwashed egg is extraordinarily resistant to bacterial penetration, the consensus of opinion is that dirty eggs, especially if washed prior to storage, are much more likely to become infected (5, 6, 9, 10). Since the washing of dirty eggs on the farm offered a possible explanation for the high count eggs the authors has encountered, arrangements were made with the Poultry Division, Central Experimental Farm, to supply naturally dirty eggs the same day they were gathered. These were then subjected to the treatments outlined below, in the hope that some light would be thrown upon the question.

EXPERIMENTAL

Three times a week, commencing May 16, 1945, one dozen "dirties" were brought to the laboratory. During the first half of these studies, alternate dozens were placed in the 37° C. incubator for 1 hour to warm up before further treatment (to facilitate bacterial penetration of the shell), while the remainder were not warmed up. From each 12 eggs, 6 were left unwashed as controls; the other 6 were washed with a wet cloth, no detergent being used. The cloth was dipped in an enamel measure containing 1500 ml. of water at around 20° C. (68° F.) before wiping the dirt, almost entirely fecal matter, from the shells. Each succeeding egg was washed in the same water, which became progressively dirtier. Without any attempt at drying, the washed eggs were returned to their cartons and, along with the controls, stored at 14° C. (58° F.) with a relative humidity of around 50%. Three weeks later they were removed; the control eggs were washed in the same way (to minimize contamination of contents during their removal from the shells), and allowed to dry. Each egg was then immersed in a 1/500 solution of Roccal at around 40° C. (104° F.) for a minute or two prior to being opened.

Using sterile forceps, an area of shell large enough to allow the egress of the contents was removed from the blunt end of the egg. The contents were then transferred to a sterile 4 oz. screw cap jar, examined for fluorescence under an EH4 mercury arc lamp equipped with a Corning No. 587 filter (8), and checked for odour and appearance. They were then rendered as homogeneous as possible[4] by means of a mechanical agitator equipped

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[4] Unpublished data indicate the difficulty of obtaining a uniform dispersion of bacteria in eggs by the methods previously employed—beating with a spoon or shaking in a jar.

with two sharp blades tilted at about a 30° angle. Plates were poured on appropriate dilutions with tryptone glucose extract milk agar (7) for total count and incubated at 30° C. for 3 days; 1 ml. portions were also plated on violet red bile agar and incubated for 20 to 24 hours at 37° C. Where the bacterial content was high, cultures of the predominant type were isolated for identification and further study.

By the time the first 4 dozen had been analysed, it became evident that very few eggs contained more than an occasional organism. (The highest count noted was 17 per ml.). Since the contamination on the shells of the eggs was almost entirely of a fecal nature, while the organisms isolated from high count eggs have been chiefly soil and water types, it was felt desirable to increase the opportunities for contamination with these types. For this purpose, surface soil was collected from in front of one of the colony houses in use in 1944; this was moistened sufficiently to make a paste, and each egg dipped into it so that approximately 25% of the shell was soiled. The entire dozen eggs were then placed in the 37° C. incubator for one hour, when the washing of one-half the lot was carried out as described.

RESULTS AND DISCUSSION

The distribution of bacteria counts and other relevant data from the variously handled lots of eggs are shown in Table 1.

The most surprising feature is the low percentage of eggs showing any appreciable number of bacteria. Bryant and Sharp (1), examining the whites and yolks of naturally dirtied eggs which had been washed, then held at room temperature for 30 days, found all the way up to 100% containing over 100 organisms per ml.; yolks of unwashed controls were infected in 83% of the cases. (Unfortunately, these workers failed to indicate the magnitude of the counts obtained on infected eggs, so that comparisons on this point cannot be made.) This is the more surprising because our treatments of the eggs, including warming before washing, are believed to be more favourable to bacterial penetration than were the methods they employed.

In our studies, supplementary soiling of the shells with mud resulted in a significant increase in the number of eggs carrying more than 250 bacteria per ml., as well as in the average count per egg. However, the latter figures are greatly influenced by a single egg which gave a count of 62,000,000 per ml.

Washing in itself had much less effect upon the number of infected eggs than had been anticipated. The number of washed eggs with counts in excess of 250 per ml. was identical with that of the unwashed controls (7 in each). However, 4 of the 7 washed eggs gave counts of 2,100,000 per ml. or higher (maximum 62,000,000), while only 1 of the controls showed a count (4,900,000) in excess of 40,000 per ml. (Table 2). The number of bacteria which would be contributed to melange by the washed eggs is therefore significantly higher than for the controls, as indicated by the average counts; 643,778 for 130 washed, and 38,638 for 129 control eggs.

Warming prior to washing also influenced the number of infected eggs less than was expected. In the case of the control eggs which did not

TABLE 1.—DISTRIBUTION OF BACTERIA COUNTS FROM 259 DIRTY EGGS WASHED BEFORE OR AFTER STORAGE FOR 3 WEEKS AT 14° C. (58° F.)

Controls	Count per ml.														
	No. of eggs	Median count	Average count	< 1	1-10	11-50	51-250	251-1000	1001-5000	5001-25,000	25,001-100,000	100,001-500,000	500,001-2,500,000	2,500,001-10,000,000	> 10,000,000
Washed after storage															
A. Fecal contamination on shell															
1. Not warmed before storage	29	< 1	1,380	17	11						1				
2. Warmed before storage	34	< 1	1	20	13	1									
B. Feces and mud on shell															
Warmed before storage	66	2	74,913	8	45	3	4	2		2	1			1	
Washed before storage															
A. Fecal contamination on shell															
1. Not warmed before storage	30	< 1	7,668	16	12	1						1			
2. Warmed before storage	36	< 1	102,786	23	9	1	2							1	
B. Feces and mud on shell															
Warmed before storage	64	1	1,255,825	14	40	2	2	1		2			1		2

TABLE 2.—DATA CONCERNING EGGS SHOWING COUNTS ABOVE 5,000 PER ML.

Egg No.	Bacteria count per ml.	Coliform organisms per ml.	Fluorescense of egg	Predominant organism isolated	Large pores in shell	Treatment of egg: Washed before storage	Washed after storage	Warmed before storage	Not warmed before storage	Soiled with feces	Soiled with mud and feces
229	5,600	< 1	–	Micrococcus			x	x			x
368	6,800	< 1	–			x		x			x
245	8,000	< 1	–				x	x			x
424	9,400	< 1	–		x	x		x			x
407	29,000	< 1	–	Escherichia	x		x	x			x
145	40,000	< 1	–	Flavobacterium	x		x		x	x	
186	230,000	< 1	–	Flavobacterium		x			x	x	
302	2,100,000	< 1	+++	Bacterium	x	x		x			x
170	3,700,000	60	–	Flavobacterium		x		x		x	
249	4,900,000	< 1	–	Pseudomonas	x		x	x			x
244	15,000,000	< 1	–	Flavobacterium	x	x		x			x
282	62,000,000	< 1	+++	Bacterium	x	x		x			x

receive a supplementary soiling with mud, the warmed eggs actually showed up better than the unwarmed. However, average counts per ml. were significantly higher for the entire lot of warmed eggs, being 441,013 for the latter (200 eggs) and only 4,577 for the 59 unwarmed eggs.

Since eggs with large pores in the shell might be expected to become infected more readily (1, 2), the presence of such pores, as indicated by the escape of air bubbles when the cold egg was immersed in warm Roccal solution, was noted when observed. While few eggs with high counts failed to show large pores, a surprising number with large pores remained virtually free from bacteria.

Examination under ultra-violet light again proved to be of limited value in the detection of high count eggs. As will be seen from Table 2, only 2 eggs showed fluorescence. Both of these gave counts in excess of 2,000,000 per ml. As in our previous studies (8), there was no close correlation between fluorescence in the egg and the presence of organisms of the genus *Pseudomonas*. Fluorescence was not noted in any of the remaining 257 eggs examined.

A word of explanation is necessary concerning Egg No. 407 in which organisms of the genus *Escherichia* were predominant, although no coliform organisms showed up on the violet red bile agar at the initial examination. The tryptone glucose agar plates poured from 1 ml. of this egg showed over 300 mould colonies; one of the moulds isolated was tested out in another connection for its antibiotic potency, and was found to exert a definite inhibitory action on *E. coli*. This may explain the inability of the coliform organisms to develop sufficiently on the violet red bile medium to be recognized after 20 to 24 hours incubation.

In view of the fact that all eggs as received were found to be smeared with fecal matter, it was surprising to find coliform organisms present in only two other eggs. One of these had been warmed and washed before

storage; it gave a count of 60 coliforms per ml. along with a total count of 3,700,000 per ml. The other, not washed until after storage, gave a count of 2 per ml. with a total count of 6 per ml.

In view of the apparently greater resistance to infection shown by the eggs in these studies, it was felt that some other factor might be involved. Since the eggs studied all came from the Central Experimental Farm flock where the hens receive a ration which is regarded as nutritionally adequate, it seemed possible that this might be a factor inasmuch as many farm flocks may not receive as adequate a ration. This point may warrant further investigation.

SUMMARY

A surprisingly small percentage of 259 new-laid dirty eggs contained appreciable numbers of bacteria after being stored at 14° C. (58° F.) for 3 weeks.

Washing with a wet cloth before storage did not increase the number of infected eggs, but the average count of such eggs was appreciably higher than that of eggs washed shortly before analysis.

Warming the egg prior to washing also influenced the number of infected eggs less than was expected, although the average count was again higher than for the unwarmed eggs.

Supplementary soiling with mud of eggs naturally soiled with fecal matter increased the number of infected eggs as well as the average count. Coliform organisms were rarely encountered.

ACKNOWLEDGMENTS

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THE RELATIVE ANTIRACHITIC POTENCY OF IRRADIATED ERGOSTEROL (D_2) AND IRRADIATED 7-DEHYDRO-CHOLESTEROL (D_3) FOR GROWING PIGS[1]

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It is commonly believed by nutritionists and feeders that swine require a dietary source of vitamin D or its equivalent in ultra-violet irradiation. Complete absence of some antirachitic agent presumably leads to faulty and/or slow bone formation demonstrable experimentally by means of size measurements, by line test, or by bone ash determinations. There is no question but that under some conditions of practical feeding and management, young pigs may become lame and clinically 'rachitic.' Furthermore, such conditions are reportedly prevented and, if not too advanced, cured by the feeding of some potent source of vitamin D.

Strangely enough, however, entirely satisfactory evidence concerning the minimum vitamin D requirements for swine is not to be found in the literature. Indeed the occurrence of clinical rickets appears to be highly variable among experimental animals confined to so called 'rachitic' diets.

One of the latest reports bearing on this question is that of Braude, Kon, and White (2) who found no rickets among pigs fed normal diets containing 0.5% P or over and a Ca : P ratio of 1.4 : 1. Similarly, Bethke *et al.* (1) found that within Ca : P ratios of 1 : 1 to 2 : 1 and with rations containing not less than 0.6% P, satisfactory growth and bone formation took place without vitamin D. Dunlop (4) concludes that diets providing 0.6% P and a Ca : P ratio of 0.75 : 1.0 do not need vitamin D supplementation. It is interesting to note the finding of Johnson and Palmer (6) that white pigs become rachitic more slowly than dark-skinned hogs because of greater storage of vitamin D prior to their experiments. This difference in skin colour was reported by Marek *et al* (cited by Braude *et al.* (2)) who also give 1.85 gms. P per 10 kgms. body weight and a Ca : P ratio of 2.2 : 1 as the minimum of P for white pigs where no vitamin D is furnished. On this basis, and figuring that a 50-pound pig requires 2.7 pounds air-dry feed, the daily P need would be 4.2 gms. or equivalent to 0.3% of the ration.

Against these findings must be set numerous reports in which rickets was found among pigs, in some cases even with apparently adequate Ca and P ratios and intakes. For example Braude *et al.* (2) found that the introduction of brewers' dried yeast into the ration brought on rickets which was not cured or prevented by extra Ca or P but responded to small doses of cod-liver oil (about 2 gm. cod-liver oil per pig per day, apparently furnishing some 200 i.u. vitamin D per pig per day). Senior (7) found rickets among young weaned pigs never exposed to sunlight or given vitamin D. This diet carried Ca/P ratios ranging from 0.76 : 1 to 1.53 : 1 and with P as 0.78% of the ration. Their dams had had access to sunlight during pregnancy and lactation. The development of the rachitic condition was prevented by additions of (about) 1% of cod-liver oil to the diet.

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It is difficult to reconcile these contradictory findings. Apparently in the absence of known sources of vitamin D, growing pigs may or may not develop a rachitic condition. Rickets appears to be a more certain result when the intake of P or of Ca falls below certain minimum values, or when the Ca/P ratio deviates widely from 1 : 1.

One factor which may be involved is growth rate. The requirements for Ca and P are obviously closely associated with the live weight gains being made, and if, through restriction of the intake of these minerals, the pigs reduce their rate of gain sufficiently, then no rickets is to be expected. It may be pointed out that 'normal' gains as frequently reported imply gains comparable to some standard. Unless the standard used is stated, the results cannot be adequately judged. In the majority of reports in which growth rates are cited, they are below the 'expected gains' set out in the Recommended Nutrient Allowances for Swine published by the Committee on Animal Nutrition of the U.S. National Research Council (9).

In view of the increasing use in farm rations of synthetic vitamin D, the question of the requirements of swine for this vitamin has again arisen, together with the added problem of whether or not the irradiation products of different sterols, particularly of ergosterol (D_2) and 7-dehydrocholesterol (D_3), are of equal effective antirachitic potency for this species.

Accordingly in 1943 a study was undertaken with the specific object of comparing the effectiveness of vitamin D_2 and D_3 in preventing rickets in pigs placed on rachitic diets at a weaning age of about 56 days. Two tests have been completed and are hereinafter reported.

EXPERIMENTAL

Test I

The prophylactic method of assay was employed, using percentage ash in moisture-fat-free bone as the criterion of the antirachitic value of the vitamin D given.

The reason for using this method, in spite of the somewhat larger numbers of animals required, was that it avoids a preliminary depletion period and the problems of establishing and identifying clinically equal degrees of rickets, procedures for which are at present not available for swine.

Thirty-two Yorkshire pigs, weaned and started on test at 56 days of age were alloted to 8 lots of 4 pigs each. The allotment was at random subject to the restriction of equal numbers of each sex in each lot of 4.

The pigs during the assay period were penned and fed individually. They were weighed and feed checked weekly.

The basal feed mixtures for all pigs consisted of:

Ground wheat	43.5 lb.
Ground oats	45.0 lb.
Dried liver and lung meal	7.0 lb.
Calcium carbonate	4.0 lb.
Salt	.5 lb.
Carotene	150.0 mg.
Vitamin B complex: Thiamin 1 part, Riboflavin 2 parts, Niacin 4 parts	315.0 mg.

Based on average samples of the above feeds this mixture carried about 15% protein, 1.66% Ca, and 0.43% P.

Experimental evidence indicates that while the most usually occurring swine rickets is of the low Ca type, a ration with between 0.30 and 0.50% P with a Ca/P ratio of about 4 may be expected in the absence of vitamin D to produce a subnormal bone ash; and that at these same intakes, normal bone ash will be found if adequate vitamin D is included in the ration.

The general design of the test is indicated by Figure 1 showing the allotment of the pigs to the several groups.

FIGURE 1.—ALLOTMENT PLAN OF VITAMIN D TEST
(Total pigs, 32)

Vitamin D per day/pig	Form of Vitamin D	
	Ergosterol (D_2)	7-dehydro-cholesterol (D_3)
Check	4 pigs	
10 i.u.		4 pigs
20 i.u.	4 pigs	
40 i.u.		4 pigs

Results of Test I

It was intended that the pigs should be fed for a period of 42 days after which they would be killed, the long bones (ulna and radius) of the front legs removed for line test and the metacarpals examined for bone ash determination. At the end of the first 6 weeks, however, there was no clinical evidence of rickets; and 3 male pigs killed[4] showed bone ash values from 62.7% to 63.4%. The feeding period was therefore continued for a second 6 weeks after which 6 additional pigs were killed. These showed bone ash values ranging from 58.5% to 61.9%; and line tests indicated normal bone formation. The remaining 19 pigs were continued on feed for a further 4 weeks after which they were slaughtered. Bone ash values ranged from 58.5% to 63.6%. Line tests showed normal calcification. No sex differences were found.

Two control pigs killed at the start of the test had bone ash of 59.1%.

The average bone ash values for the groups are shown in Table 1.

TABLE 1.—AVERAGE % BONE ASH VALUES

Vitamin level	Form of vitamins	
	D_2	D_3
Check	61.9	61.9
10 i.u./day	61.2	61.7
20 i.u./day	61.4	60.2
40 i.u./day	59.8	58.5

[4] We are indebted to Canada Packers for killing facilities and for absorbing losses due to special cutting for all hogs in these studies.

It is obvious from the figures in Table 1 that there was no rickets in any of the groups, which of course precludes any conclusion as to the relative potency of ergosterol vs. 7-dehydrocholesterol as sources of the antirachitic vitamin. Actually the standard deviation of the % ash values was 1.4, necessitating a difference of 2.23 units of per cent between groups of 4 pigs to cover variations not related to experimental treatments. Between D_2 and D_3 there was no case of a significant difference; and the lowest values for the highest daily intakes of the vitamin, though in one case statistically significant, are biologically contrary to expectation.

The most probable explanation of the failure of rickets development was the slow rate of growth shown by all pigs. These are summarized in Table 2.

TABLE 2.—AVERAGE DAILY GAIN AND FEED INTAKE OF PIGS

Group	Initial weight	6 Weeks			16 Weeks		
		Gain	Feed	F/G	Gain	Feed	F/G
	lb.	lb.	lb.	lb.	lb.	lb.	lb.
Check	26	0.69	2.6	3.8	1.12	4.4	3.9
10 i.u.	30	.46	2.2	4.9	0.92	3.8	4.2
20 i.u.	30	.59	2.5	4.2	.94	.8	4.0
40 i.u.	24	.52	2.1	4.0	.86	3.5	4.1
D_2 all lots	27	0.48	2.1	4.4	0.87	3.5	4.1
D_3 all lots	28	.55	2.3	4.2	.95	3.9	4.2

The average gains for all pigs were 0.48, and 0.90 lb. per day during the 6- and 16-week periods, respectively. Normal gains should be nearly double these figures.

The failure of the pigs to gain normally might be charged to inadequate levels of vitamin D were it not for the fact that there was no evidence of increased gain with increasing level of vitamin D intake. Feed consumption was low and the feed required for one pound gain was higher than normal. The vitamin A intake was 50% above the requirements set out in the feeding standard proposed by the U.S. National Research Council Committee on Animal Nutrition (9).

These facts lead to the belief that the limiting factor in this diet for the growth of pigs was protein. Restriction in amount or in quality of protein normally results in low feed intake, poor feed utilization, and in slow gains. In order to prepare a ration as low in P as possible, the usual protein supplements were of necessity avoided. Liver and lung meal was chosen as one of the few high protein feeds with low P content.

Conclusions from Test I

Obviously this trial yields no data upon which conclusions relative to the usefulness of vitamin D_2 compared to D_3 as antirachitic agents in swine rations can be made. It seems evident that on rations which do not permit more rapid growth than observed in this test, rickets may not develop even in the absence of known sources of antirachitic substances in the diet and with the Ca/P ratios as high as 4 : 1.

Test II

Because of the difficulties experienced in the previous test with a suitable source of protein in preparing low P diets for swine, it was decided to employ a low Ca, high P diet in this trial. Senior (8) has reported that neither ratio of Ca to P, nor level of intake of these elements will eliminate the need for vitamin D in the swine ration. Rickets was produced in his test on diets supplying 0.6% P and 0.47% Ca. A diet containing this quantity of P will permit the use of foods normally found in swine rations which are expected to produce normal growth.

Thirty-two Yorkshire pigs weaned at 52 to 56 days of age and started on test at initial weights ranging from 17 to 28 lb. at ages of about 56 days were allotted to 8 groups of 4 pigs each. The allotment was random, subject to the restriction of equal sexes in each group.

Each group of 4 pigs was penned together and feed records made for the groups only. Feed was allowed to limit of appetite. It was prepared for feeding by pouring over the dry meal allowance, in the trough, water equal to about 3 times the weight of dry meal. The length of the feeding period was 6 weeks for all pigs, at which time one-half were killed for bone ash assay. The remaining 16 pigs were carried for a further period of 10 weeks.

The feed mixture finally used was made up as follows:

Ground barley	79.5 lb.
Linseed oilmeal	5.0 lb.
Soybean oilmeal	5.0 lb.
Defatted wheat germ	5.0 lb.
Fish meal	5.0 lb.
Salt	0.5 lb.

To each 100 pounds of the above was added:—

Ferrous sulphate	7 grams
Carotene	50 milligrams
Riboflavin	10 milligrams
Pantothenic acid	100 milligrams

Based on average samples this combination carried approximately 18% of crude protein, 0.57% of P, and 0.28% of Ca.

The allotment plan and the levels of vitamin D supplements were identical with that shown in Figure 1 for Test I.

Examination of the complete data of this trial reveals no difference in gains between control lots and any of the 3 levels of vitamin D, and obviously therefore, none between the two forms of vitamin D used.

A comparison of the average gains of all pigs as compared to the Macdonald College Standard (3) is interesting and perhaps significant in this respect.

In interpreting the curves in Figure 2 it is to be noted that the pigs which were killed after the first 6 weeks were the largest pigs in each group. These were chosen deliberately to see whether or not the most rapidly growing animals had become rachitic.

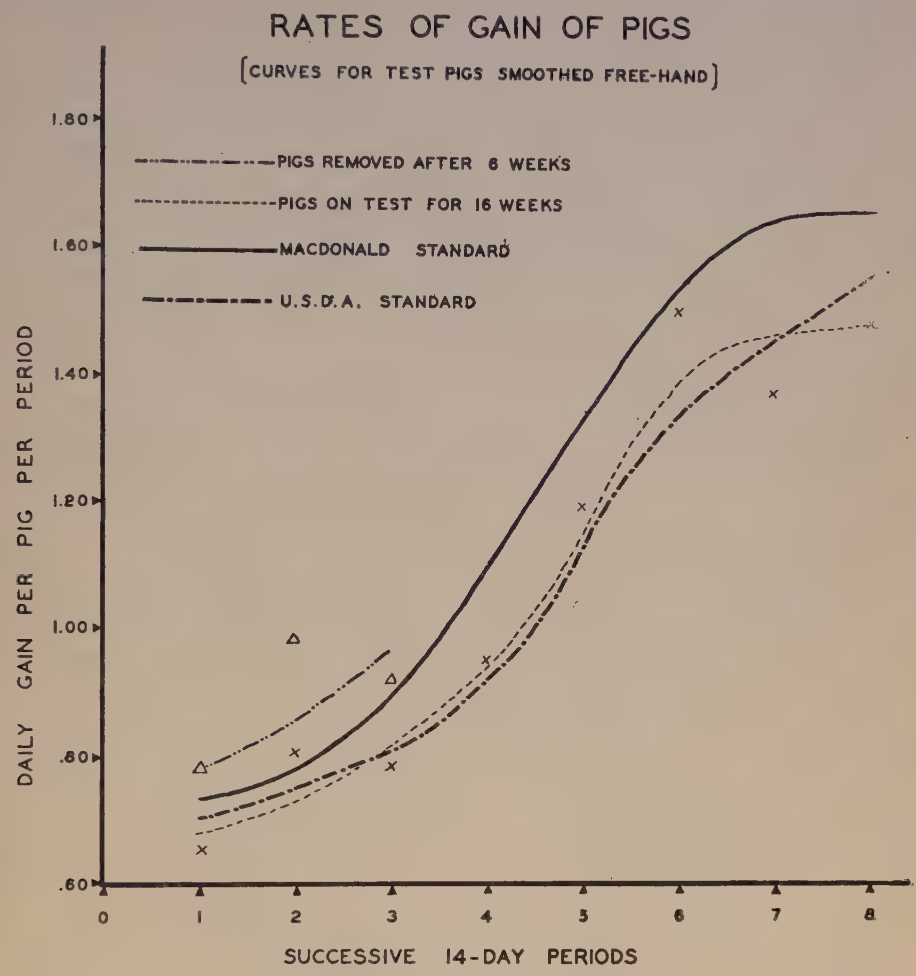

FIGURE 2. Rates of gain of pigs.

The curve of daily gain for those pigs which remained on test for 16 weeks was below the Macdonald College Standard but follows with surprisingly good fit, one calculated from the growth curves reported by the United States Department of Agriculture (5). If the gains of the faster growing pigs from this test are averaged with those of the slower half (for the first 6 weeks) their mean gains are almost exactly those expected on the basis of the Macdonald Normal Rate of Gain Curve.

The mean daily gains of all pigs for the two weeks during which they became 50 pounds in weight was 0.94 pounds which may be compared with expected gains, of 50-pound pigs, of 0.90 pounds according to the Standard of the Committee on Animal Nutrition of the United States National Research Council.

In so far as rickets is concerned, it failed to develop in a single pig. In Table 3 are given the mean gains and bone ash values obtained from this test.

TABLE 3.—AVERAGE GAINS AND BONE ASH VALUES ACCORDING TO SOURCE AND AMOUNT OF VITAMIN D FED

Dose level	All pigs—First 6 weeks				Pigs remaining on test 16 weeks			
	Irradiated Ergosterol (D_2)		Irradiated 7-Dehydro-cholesterol (D_3)		Irradiated Ergosterol (D_2)		Irradiated 7-Dehydro-cholesterol (D_3)	
	Gain 6 wk.	Bone ash	Gain 6 wk.	Bone ash	Gain 16 wk.	Bone ash	Gain 16 wk.	Bone ash
	lb.	%	lb.	%	lb.	%	lb.	%
Check	34	57	34	58	121	58	121	59
10 i.u./day	39	57	32	57	137	57	107	58
20 i.u./day	37	59	30	57	157†	57	101	58
40 i.u./day	35	56	38	59	120	55	132	54
Mean gains, all levels of Vitamin D	37	57	33	58	134	56	113	57
Normal gain, Macdonald College Standard	34 lb.				136 lb.			

* Average of 2 pigs per lot killed after 6 wk. feeding. Based on moisture-fat-free metatarsals.
† One pig only. Mate died from causes not related to test.

Chick Assay in Connection with Test II

In view of the complete absence of rickets, it was thought desirable to examine, as a possible source of vitamin D, the fish meal used, since it contained 16% fat (ether extract). Accordingly an assay[5] was arranged in which 4 groups of 20 chicks each were fed on a ration composed of:—

Yellow corn	58 lb.
Wheat middlings	25 lb.
Fish meal	12 lb.
Yeast	2 lb.
Precipitated tricalcium phosphate	2 lb.
Salt	1 lb.
$MnSO_4 \cdot 4H_2O$	10 grams

The fish meal in the above mixture was prepared as follows:—

Group	Extracted fish meal	Unextracted fish meal	Potential A.O.A.C. Units D/lb. ration[6]
	%	%	
Lot I (control)	100.0	0.0	0
Lot II	97.5	2.5	11
Lot III	95.0	5.0	22
Lot IV	90.0	10.0	44

[5] The co-operation of the Department of Poultry Husbandry is gladly acknowledged in this assay.

[6] The proportions of extracted and unextracted fish meal were such that on an assumed vitamin D potency of 50 A.O.A.C. units per gram for the oil in the fish meal, the diet of Lot IV would carry about 44 A.O.A.C. units of D per pound of ration as fed.

The chicks were confined to the test diets for 21 days, after which bone ash values of the leg bones were determined. The results were as follows:—

Group	Ash of Moisture-fat-free bone
	%
Lot I (control)	32.2
Lot II	33.4
Lot III	32.1
Lot IV	32.1
Normal	45.0

These data fail to indicate any measurable antirachitic potency of the fish meal used. Unless the fish fat contained principally D_2, which seems unlikely, it could be presumed that the fish meal was devoid of substances antirachitic for pigs.

DISCUSSION

These tests offer no basis for a comparison of the antirachitic potency for swine of vitamin D_2 or D_3. Indeed it would seem that the need of young growing pigs, from weaning time to weights of 175 pounds, raised entirely indoors during winter, is less than has been commonly believed. The pigs were from dams that received daily liberal amounts (8cc. of 2,000 A — 400 D) of feeding fish oil during pregnancy and lactation. During lactation (56 days) the fish oil allowance was put into the sows' feed troughs once daily. The litters had access to this feed and undoubtedly ate varying amounts of it. If they obtained enough of the vitamin D to prevent rickets during the next 16 weeks, then the storage of this vitamin in the body is more extensive than was found by Senior (7). Our results in this respect, however, are in agreement with the findings of Braude *et al.* (2) in so far as any need for vitamin D to prevent rickets is concerned.

It is to be recognized that in neither of these trials did the pigs gain as rapidly as called for by our own (Macdonald) standard. In the first trial this, we believe, can be accounted for by poor quality protein. In the second test it will be remembered that after 6 weeks, 2 pigs in each group of 4 pigs, were removed for determination of bone ash. The most rapidly growing pigs were deliberately chosen, thus leaving the slower growing pigs for the balance of the trial. The average gains, using all pigs up to 6 weeks, were normal according to the Macdonald College Standard (see Figure 2), and since the trend of gains of the pigs left on test parallels that of our standard it seems unlikely that their somewhat (0.15 lb. per day) slower growth rate was traceable to vitamin D deficiency. In any case it cannot be assumed that the failure of rickets was due to slow growth.

CONCLUSIONS

1. These tests yield no data on which to judge the relative antirachitic effectiveness of irradiated ergosterol (D_2) and irradiated 7-dehydrocholesterol (D_3) for young growing pigs.

2. Weaned at 8 weeks from mothers, who during pregnancy and lactation have received daily about 15,000 i.u. vitamin A and 3,000 i.u. vitamin D, young pigs confined indoors appear to have a considerably lower vitamin D requirement than present standards call for if:

(1) their diets contain at least 0.57% P;

(2) the Ca/P ratio is not wider than 0.5 : 1;

(3) bone ash values of the order of 57%, and normal trabecula, are taken as indicating normal bone metabolism.

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TESTING WHEAT SEEDLINGS FOR RESISTANCE TO HELMINTHOSPORIUM SATIVUM[1]

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INTRODUCTION

In Western Canada, plant breeders have made great advances in developing better cereal varieties, especially wheats resistant to stem rust. Occasionally a breeding program is well underway when the appearance of some unexpected disease may cause serious damage in the nursery and necessitate a change in plans. Sporadic infestations by common root rot fungi, notably *Helminthosporium sativum* P. K. & B. and *Fusarium* spp., in plant-breeding nurseries have been brought to the attention of pathologists from time to time. Studies on root rots, including varietal resistance, have been conducted for many years. It is generally agreed that the organisms mentioned above are usually associated with all outbreaks of common root rot and their pathogenicity can be readily demonstrated. Nevertheless the etiology of root diseases is complex, and the role of other fungi as well as the influence of the environment must not be overlooked. A step forward, however, will have been made if reliable and quick methods can be developed to analyse the reactions of a large number of varieties and lines to the well-known pathogens. This investigation, therefore, was confined almost entirely to the reaction of wheat seedlings to inoculation with *H. sativum;* in addition, strict attention was given to ways and means by which time and space might be conserved.

The plant breeder must finally test his material in plot and field to determine vigour, yield, and general quality. Such tests over a period of years and in many districts soon eliminate the weak lines. Presumably, selection has also eliminated those sorts most susceptible to root diseases. It would appear that something like this has functioned in the development of our better wheat varieties, for on the whole they have been vigorous and high yielding. However, the sporadic occurrence of root diseases as well as the possible occurrence of physiologic races makes this procedure very hazardous. A better knowledge of root diseases obtained through improved techniques should greatly reduce the trial and error effort.

The root rots were not well understood when expansive plant breeding programs were commenced in Western Canada about 1925. Three types of root disease were then under study. Investigations on resistance included inoculation tests in the greenhouse and field, the establishment or attempted establishment of root disease gardens by repeated soil inoculations, and observations of natural infections in comparative trials. Most of these studies gave fairly good comparative data but they all failed to provide advance information to the plant breeder. Such studies added little to what the breeder would obtain in the course of his ordinary field

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experiments. Some of the methods used and results obtained in Western Canada have been reported by Greaney, Machacek, and Johnston (2) and Tyner and Broadfoot (6). Studies on varietal resistance to common root rot have been conducted in this laboratory for many years. Some of the earlier tests were made by Mr. G. A. Scott while in later years the investigations were continued by Dr. B. J. Sallans. Rather extensive greenhouse and field experiments were reported in 1926 (4) and 1927 (5).

This and subsequent work as well as co-operate studies with Dr. J. B. Harrington at the University of Saskatchewan impressed upon us the necessity of more investigations on techniques. Those who have studied root diseases have noticed and reported from time to time upon tests dealing with varietal differences. The early work in this connection was reviewed by Christensen (1) who also gave the results of his extensive experiments with wheat varieties. Although all of this research has been valuable and helpful to the plant breeder, there still exists a great need for simple methods which would enable him to assess parental material and analyse resultant lines.

MATERIAL AND METHODS

The common root rot pathogen *Helminthosporium sativum* was used in most of the tests although a few trials were run with *Fusarium culmorum* (W. G. Sm.) Sacc. Both fungi has been in the laboratory stock for some time, but only freshly transferred cultures were used in these experiments. Many new isolates of *H. sativum* were made in 1944, and some of these were compared for virulence. In the preliminary tests, seed of the ordinary wheat varieties was used while later, various species, varieties, and lines were employed.

All of the first tests were made in test-tubes or culture-tubes and may be referred to as the test-tube test. Under certain conditions, *Petri* dishes fitted with a moistened filter paper were useful. In the test-tube method, the tube is fitted with a strip of blotter paper wide enough to fit nicely in the tube and of about the same length as the tube. The top portion of this strip is bent to form a shelf about two inches from the top of the tube (Figure 1). Water is added to the level of this shelf. In handling a large number of tubes, which are first placed in racks, it was found most convenient to fill all tubes to the top; then with a small pipette attached to a suction-pump; the excess water is drawn off and the shelf properly adjusted. The racks are now ready for seeding. The seeds are inoculated with a conidial suspension, usually from a 7-day old slant culture strained to remove large fragments. The suspension is poured over the seeds in a syracuse watch glass, then with forceps they are picked up and placed on the blotter shelf. The racks may then be placed in an incubator or kept in the laboratory, preferably the former. In 7 days, at 24° C., readings are made for degree of coleoptile lesioning, blighted seed, non-germination, etc. Generally, most attention was given to coleoptile lesions. Checks of uninoculated seed were run for viability, lesions, mouldiness, and so on.

In the Petri dish method, a larger number of seed may be used with greater economy of space; although this method cannot be employed if the seed shows a tendency to mould. A few mouldy kernels soon spoils a

test. This was one of the main reasons for development of the test-tube method wherein each seed is grown separately. Petri dishes are prepared by placing a filter paper to fit the bottom of the dish; 2 or 3 cc. of water are added at the start and another 2 cc. on the third day. The seeds are inoculated as described above and placed in the dish at the rate of 25 to a dish. This was the ordinary procedure, although in some tests the seeds were inoculated after being placed in the dish. Readings of symptoms as described above are usually possible on the fifth day.

EXPERIMENTAL RESULTS

Experiment 1

Over a period of about two years, the senior author conducted routine pathogenicity tests on several varieties of wheat. This was done with the object of studying methods. From this work, the test-tube procedure was devised. When this method was repeated from time to time, consistent varietal differences were noted in the replicates. After consultation with the junior author, the work was extended and additional attention paid to the subject of varietal resistance. In the preliminary tests the common varieties were tried using seed from the laboratory plots. The seed of Pentad, McMurachy's selection, Pelissier, and Mindum were several years old. From the Dominion Laboratory of Plant Pathology, Winnipeg, were obtained seed of Prelude, R.L. 25; H-44-24, R.L. 229; Thatcher (Double Cross), R.L. 1945; and a Marquis × Kanred hybrid. These latter sorts were studied because they represent parental material of some of the newer varieties. The seeds were inoculated with *H. sativum* as described above, and the racks were placed in an incubator at 24° for 7 days. In rating disease, most attention was given to the coleoptile lesions as they are the most distinctive symptom. For comparative purposes, the degree of infection was determined by deducting natural infections as shown by the checks from the total coleoptile lesions shown by the inoculated seed. A summary of the results of many replicates is given in Table 1.

TABLE 1.—A COMPARISON OF WHEAT VARIETIES IN THE SEEDLING STAGE FOR RESISTANCE TO *H. sativum*

Variety	Treatment	Total seeds	Seedlings			Infection
			Total	Clean	Coleoptile lesions	
				%	%	%
Marquis	Inoc.	216	178	86.4	14.6	9.6
	Check	216	179	95.0	5.0	—
Thatcher	Inoc.	216	202	73.8	26.2	18.6
	Check	216	210	92.4	7.6	—
Double cross	Inoc.	144	121	79.4	20.6	20.6*
Red Bobs	Inoc.	216	199	73.9	26.1	21.0
	Check	216	194	94.9	5.1	—
H-44-24	Inoc.	144	131	72.6	27.4	27.4*
Apex	Inoc.	216	175	67.5	32.5	30.5
	Check	216	186	98.0	2.0	—

* Check data were not available for these varieties.

TABLE 1.—A COMPARISON OF WHEAT VARIETIES IN THE SEEDLING STAGE FOR RESISTANCE TO *H. sativum*—*Continued*

Variety	Treatment	Total seeds	Seedlings			Infection
			Total	Clean	Coleoptile lesions	
				%	%	%
Marquis × Kanred	Inoc.	144	104	53.9	46.1	46.1*
Ceres	Inoc.	192	152	41.0	59.0	57.3
	Check	144	116	98.3	1.7	—
Renown	Inoc.	288	179	38.5	61.5	57.4
	Check	144	96	59.9	4.1	—
Prelude	Inoc.	144	77	38.9	61.1	61.1*
Reliance	Inoc.	288	173	34.6	65.4	63.7
	Check	144	114	98.3	1.7	—
Pelissier	Inoc.	228	108	12.9	87.1	78.2
	Check	144	89	91.1	8.9	—
McMurachy	Inoc.	216	121	12.3	87.7	78.7
	Check	216	165	91.0	9.0	—
Reward	Inoc.	228	160	9.3	90.7	79.5
	Check	144	107	88.8	11.2	—
Pentad	Inoc.	228	117	16.2	83.8	80.9
	Check	144	101	97.1	2.9	—
Regent	Inoc.	216	169	7.0	93.0	83.0
	Check	216	183	90.0	10.0	—
Mindum	Inoc.	228	104	9.6	90.4	88.3
	Check	144	95	97.9	2.1	—

* Check data were not available for these varieties.

Note.—The infection is the difference in per cent between lesioned seedlings of inoculated and check.

The results are considered indicative of possible trends but by no means final. Past experience with this type of infection had revealed its great sensitivity to its environment, with a subsequent variable expression of lesions. The rather consistent agreement between the many replicates, however, added greatly to the value of the ratings. Then again only seedlings were considered, and these were classed as having clean coleoptiles or coleoptiles with distinct lesions of the type caused by *H. sativum*. Some varieties showed a large number of blighted seed, obviously invaded and quickly killed. These and other signs and symptoms, although of some interpretive value, were not considered in the early trials. On the basis of the ratings in this experiment, Marquis was the most resistant variety. In most of the tests, it had shown not more than a moderate amount of infection. Thatcher was next to Marquis in resistance in these tests. On the whole, Thatcher was the most consistently resistant wheat studied. In the top group, it should be noticed, we have three well-known varieties that have shown, over a period of years, good all-round performance; namely, Marquis, Thatcher, and Red Bobs. Their resistance to *H. sativum* and

possibly other root pathogens might well explain their acceptance by farmers who must base their judgment mostly on yield and quality. We have no information on parental material of Red Bobs and Marquis except one small test with Red Fife in which it showed moderate to low susceptibility. In the Thatcher pedigree, there are Marquis, Kanred, and Iumillo wheats. The first two appear to have fair to good resistance as shown by seedling tests, while Iumillo was classed as poor in one trial.

When the new Canadian wheats are considered, Apex is the most resistant. Apex came from a cross between (H-44-24 × Double Cross) and Marquis. These parental lines all showed up well in this test. It should be mentioned that the Double Cross hybrid line used here certainly was of the same Thatcher origin and closely related to if not the same hybrid as used in the original Apex cross. The other two well-known Canadian varieties, Renown and Regent, showed little resistance to *H. sativum* and they both have Reward, a highly susceptible variety, in their pedigrees.

The two durum varieties, Pelissier and Mindum, were very susceptible; this is in agreement with general observations.

The quality of the seed is thought to be quite important in tests of this nature. There was considerable non-germination in some wheats, particularly with old seed. This factor, which was further aggravated by mouldiness in some and an increase in blighting when inoculated, made it difficult to assess this portion of the results. This, however, may be overcome by devising a disease rating formula.

Experiment 2

This test was run to compare some varieties having seed of uniform quality. Thatcher, Marquis, Apex, and Reward were grown in the greenhouse and examined at heading time for trueness to type by J. Whitehouse, Field Husbandry Department, University of Saskatchewan. The seed was of high viability. The inoculations and procedures were the same as in the above experiment with the disease rate being taken as the percentage of seedlings with coleoptile lesions. Checks were not used. As a rule, greenhouse seed is quite free of infections of the type studied here.

TABLE 2.—A COMPARISON OF WHEAT VARIETIES IN THE SEEDLING STAGE FOR RESISTANCE TO *H. sativum* USING GREENHOUSE GROWN SEED

Variety	No. of seed inoculated	Seedlings	
		Clean	Diseased
		%	%
Thatcher	100	76	24
Marquis	100	63	37
Apex	100	54	46
Reward	100	11	89

The results revealed the usual arrangement of these varieties, with Thatcher the most resistant, Reward the least, and Marquis and Apex intermediate.

Experiment 3

This experiment with three varieties will serve to show results with Petri dishes and also how a disease rating formula may be applied. As a rule, 25 seeds are sown to a dish after inoculation and each dish serves as a replicate. The material can usually be read on the fifth day, when the seedlings are classified and valued as follows: clean, 0; coleoptile lesions, lesions, trace 1, slight 2, moderate 3, and severe 4. The disease rate is derived by a formula originally used by McKinney (3), as follows:

$$\frac{\text{Sum of all numerical ratings}}{\text{Total number of seedlings with lesions} \times \text{maximum rate}} \times 100 = \text{Disease rate.}$$

Applying this to the first line of data in Table 3, we have:

$$\frac{5 + 20 + 9 + 8}{23 \times 4} \times 100 = 45.6$$

The results for this experiment are shown in Table 3.

TABLE 3.—A COMPARISON OF WHEAT VARIETIES IN THE SEEDLING STAGE FOR RESISTANCE TO *H. sativum* USING A PETRI DISH MOIST CHAMBER METHOD

Variety	Replicate	No. of seed	Clean seedlings	Coleoptile lesions				Disease rate	
				Trace	Slight	Moderate	Severe	Replicate	Average
Thatcher	1	25	3	5	10	3	2	45.6	
	2	25	2	7	5	4	6	55.2	
	3	25	1	8	11	0	3	45.6	48.4
	4	25	2	8	4	2	4	47.5	
Reward	1	25	0	6	3	8	6	65.2	
	2	25	0	4	4	2	9	71.0	
	3	25	0	3	8	5	3	60.5	60.6
	4	25	6	4	7	2	5	45.8	
Little Club	1	25	0	10	5	4	4	52.1	
	2	25	0	5	5	5	8	67.4	62.8
	3	25	1	5	3	7	8	66.6	
	4	25	1	7	3	4	9	63.5	

This method is quite satisfactory if rapidly spreading moulds do not appear. It has the advantage of using little space for large populations.

A disease rate based upon weighted degrees of lesions tends towards a fairer evaluation on border-line cases. It can, of course, be applied to the test-tube method as well.

The variety Little Club did not do well in this test although in other trials with different seed it proved fairly resistant. This variety is worthy of more attention from a root disease standpoint.

FIGURE 1. The test-tube method showing a wheat seedling after 7 days' growth at 24° C. The seed rests on a blotter paper strip which serves also as a wick. Shoot and root growth are readily observed.

DISCUSSION

It was not the main purpose of this study to delve into or review the work on the causes of resistance to this type of disease. The literature mentioned and our own work heretofore show quite definitely the existence of varietal differences to invasions by *H. sativum*. It appears that this pathogen is one and possibly the principal pathogen in causing common root rot of wheat in Western Canada. Therefore, tests with this fungus are of direct value in any wheat breeding program. As mentioned before, differences between varieties have been determined by many workers, but

usually these determinations are made from field or greenhouse tests, involving much time and labour. Frequently, the trials are made with varieties or lines which have already survived extensive field tests in the plant breeder's nursery, where the more susceptible or those of low root vigour presumably would be eliminated. Very often, therefore, the pathologist is studying lines which have been exposed to and have survived various root disease milieux. Great differences would not be expected in such material, and this appears to be the case in testing many of the new varieties. Such a procedure, although satisfactory for a long-time arduous program, certainly does not provide a quick and easy way of recognizing and classifying material suitable for parent stock. Investigations, therefore, must continue for reliable and rapid analytical methods by which large quantities of prospective parental stocks as well as new lines may be tested. It is encouraging to note in general that the results of our tests agree well with our field observations over the past few years, especially in regard to the resistance of Thatcher and the susceptibility of Reward.

The tests studied here appear to have some promise. On the whole the test-tube method is more reliable for the general run of seeds where moulds may be a disturbing factor. It is suitable for germination tests from which one can readily determine natural infections, mouldiness, and vigour; all of which is necessary information before one can evaulate inoculation results. The Petri dish method is certainly economical on space and with mould-free seed should be satisfactory. It is desirable to conduct all tests at a known suitable temperature such as 24° C. This angle, however, should be studied further. The same is true regarding whether the seedlings should be incubated in the dark or light. In so far as these factors were observed, there did not appear to be any great difference.

There seemed to be no doubt that seed to be tested should be of high viability and of good general quality. This is particularly true for the Petri dish method. In fact, it is our opinion that the seed of all material to be compared should preferably have been grown under the same conditions.

Besides the varieties mentioned above, numerous small tests both in tubes and plates were conducted with other varieties and cereals. Preliminary trials were also carried out on a large number of lines obtained from plant breeders. In many cases, distinct and consistent differences were noted. A few inoculations with *Fusarium culmorum* showed that it could be used in the methods outlined above. Tests were run with isolates of *H. sativum*, and although there did not appear to be sharp differences, there was sufficient variation to warrant additional observations.

It is our hope that rapid methods may help in analysing plant breeding material. Furthermore the test-tube method seems to have some value for germination tests, disease examination, and pathogenicity, vigour, and nutritional studies.

SUMMARY

1. The results of preliminary studies on rapid methods for testing wheat varieties for resistance in the seedling stage to infections by *H. sativum* are given.

2. Two methods were used. The first consists of test-tubes fitted with a strip of blotter paper folded to provide a shelf about 2 inches from the top of the tube. Water is added level with the shelf. The inoculated seeds are placed on the shelf. After 7 days of incubation, the infections are recorded. In the second method, Petri dishes fitted with moistened filter paper are employed. The inoculated seeds are laid in rows and incubated for 5 days.

3. The tests show that Marquis, Thatcher, Red Bobs, and Apex have more resistance than Mindum, Regent, Reward, Pelissier, and Renown. The trials were with seedlings only; and although possibly of value in analysing breeding material, any interpretation of the behaviour of a variety beyond the seedling stage must be made with caution.

ACKNOWLEDGMENT

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THE EFFECT OF FREEZING AND COLD STORAGE UPON THE BACTERIAL CONTENT OF EGG MELANGE[1]

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The effect of freezing and subsequent storage upon the bacterial content of egg melange has been studied by various workers. Stiles and Bates (16) reported a definite increase in count during the early part of the storage period; melange from strictly fresh eggs increased in count up to 3 months, then declined. Swenson and James (17) showed a drop in count from 150,000 to 60,000 per gram following freezing in dry ice and storage for 8 months at −20° C. (−4° F.). Nielsen and Garnatz (13) reported sharp reductions in counts of whole eggs containing 14% added salt after 41 days storage at −18° C. (−0.4° F.); yolks with 10% added sugar, on the other hand, showed an increase in the 20° C. count after 36 days, with a subsequent decline on further storage. Holtman (3) reported a 99% reduction in total numbers of bacteria and in most instances absence of coliform types after 7 to 9 months storage at −5° to 0° C. (23° to 32° F.) Schneiter, Bartram and Lepper (15) generally found a decrease in count on resampling after 60 hours in the freezer, but in one pack the count increased from 10,000 to 62,000 per gram. Lepper, Bartram and Hillig (11) also found an occasional sample with a higher count after freezing, although the count generally declined.

From the above review, it is difficult to decide just what effect freezing may be expected to have upon the bacterial content of whole eggs. The studies reported in this paper were initiated in the hope of providing a more definite answer.

EXPERIMENTAL

In the first series, sets of 4 samples of melange from freshly filled oblong metal moulds, lined with wax paper, were taken at intervals at a local breaking plant. The moulds were placed in a sharp freezer at approximately −19° C. (−2° F.); 2 days later the frozen blocks (6″ × 7″ × 24″) were bored with a sterilized 1″ auger at the center, near the end, and midway between the first two. A composite sample from the three borings was taken for analysis. Additional samples were taken from the same block after 1, 3 and 6 months storage at approximately −15° C. (5° F.). All samples were brought to the laboratory and analyzed with a minimum of delay. Except where specified, the methods used in official control of Canadian whole egg powder (6) were employed.

In the second series, 20 samples of melange were taken from freshly filled moulds of the same type and size at an Eastern Ontario breaking room, and analyzed at once in the plant laboratory. After 44 hours in a sharp freezer at −21° C. (−5° F.), the frozen blocks[4] were bored as described for the first series, and the samples analyzed without delay.

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[4] The melange represented by Sample No. 14 was spilt while being transferred to the sharp freezer, hence no sample of the frozen melange could be obtained.

Since it is well known that many of the organisms in egg melange fail to grow well at 37° C. (7, 13, 15), plates were incubated at 30° C. for 3 days. To obtain some indication of the comparative counts, additional sets of plates from the fresh melange, and from the frozen melange 48 hours later, were incubated at 37° C. for 2 days.

Smears for direct microscopic counts were prepared from the 1:10 dilution of melange, 0.01 ml. being spread over a circular area of 1 cm.^{2}. Those from Series I were stained for 15 to 30 seconds with Gray's (2) stain diluted with 2 parts of water, those from Series II with North's stain (8).

Coliform organisms were estimated using brilliant green bile broth (6), and positive tubes showing black and metallic colonies were considered as containing *Escherichia coli.*

To obtain some idea of the effect of freezing on the flora of the melange, from 70 to 84 colonies were picked from entire plates or segments of plates poured for each of the 4 fresh samples in Lot C (June 7) Series I. This was repeated when re-sampling after storage for 6 months. Cultures were purified and, on the basis of macroscopic and microscopic appearance, supplemented by physiological characteristics, placed in their respective genera.

RESULTS

For Series I plate counts for individual samples are shown in Table 1, and direct microscopic counts in Table 2. The general effect can more readily be grasped from Figure 1, where the average values for all samples in Lots B and F are shown graphically.

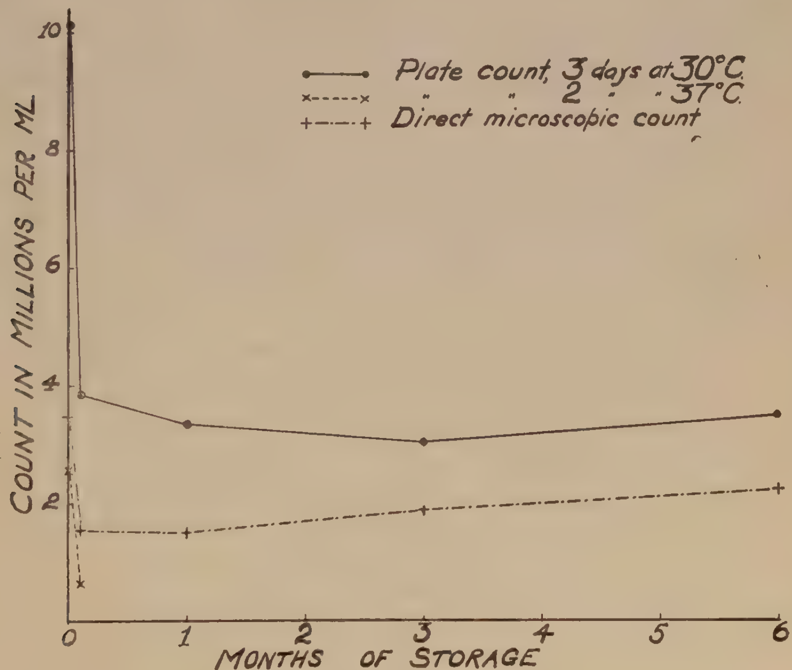

FIGURE 1.—Effect of freezing and subsequent storage on bacterial content of egg melange. (Average of 20 samples).

The results of coliform and *E. coli* determinations for Lot C are shown in Table 3. For Lot A, the dilutions employed were too high, and for the remainder too low, to permit satisfactory calculation of the most probable numbers present, hence data for these lots have been omitted.

The results of the studies on flora of fresh and frozen melange are summarized in Table 4.

For Series II, plate and microscopic counts on fresh and frozen melange are shown in Table 5, together with the percentage survival for each individual sample.

DISCUSSION

The results from Series I show an average reduction in the 30° C. plate count of nearly two-thirds as a result of freezing. On subsequent storage for periods up to 6 months, further changes were slight. The reduction on individual samples in this series ranged from 13 to 87%, with an average of 64.2%. In Series II (Table 5) the average reduction was of the same magnitude (61.66%) but there was less variation between individual samples, values for which ranged from 41.5 to 73.3% reduction. These results are in contrast to those of Holtman (3), who found a 99% reduction in total count after 7 to 9 months storage in a frozen condition (−5° to 0° C. (23° to 32° F.)). This greater reduction may be due to the higher storage temperature; Jensen (5) reported "destruction of bacteria in frozen-egg magma . . . . . . is most rapid when the products are stored at −6.7 to −3.9° C. (20° to 25° F.)."

That a similar reduction in count takes place when melange of lower initial bacterial content is frozen is indicated by data recently furnished by Mr. E. W. Noton, resident inspector for the Special Products Board at Winnipeg. Cartons containing 7 oz. of freshly prepared melange were held in a freezer at −15° to −6° F. (−26° to −21° C.) from June 12 to June 27, 1945, when a chip of frozen melange was removed from each carton, defrosted and analyzed. The remaining contents were then defrosted at room temperature (74° F.), taking 5 hours to defrost completely. After thorough stirring with a sterilized spoon, a second analysis was conducted. Bacteria counts were estimated by means of the Burri slant technique (9), slants being incubated at room temperature for 3 days. The results (Table 6) indicate a reduction in count comparable to that obtained with high count melange by the plate count method (Table 1). The data in the final column, representing analyses of the entire contents of the cartons on defrosting, suggest that some growth took place during defrosting of the remaining contents.

As anticipated, the counts at 37° for 2 days were much lower than those at 30° for 3 days. The average initial 37° count was 25.4% of the 30° count, while that after 48 hours freezing was 21.9% of the 30° count at that time.

As might be expected from the irregular distribution of bacteria in melange (7) the microscopic counts (Table 2) showed considerably more variability than did the plate counts (Table 1). Surprisingly, the microscopic counts were, with rare exceptions, considerably lower than the 30° plate counts, although generally higher than the 37° counts (Figure 1).

Lower microscopic counts than 30° plate counts were also encountered in Series II (Table 5), although the percentage surviving freezing was much higher than that indicated by the plate count. While a few of the samples reported on by Lepper, Bartram and Hillig (11) showed lower microscopic counts, in most instances the plate counts after 72 hours at 32° C. were lower both before and after freezing. Our results with fresh and frozen melange are in sharp contrast to those obtained with whole egg powder (6, 8, 11) using North's stain and incubation of plates at 37° C. for 48 hours. (With powder, plate counts at 30° in 1943 were comparable to those at 37°, although significantly higher 30° counts were obtained in some recent studies (7)). These results suggest either that certain bacteria in melange fail to stain, or else that some are lost from the smear during defatting, fixing, staining and washing. The irregular distribution of bacteria in melange, previously mentioned, scarcely affords an adequate explanation of the generally lower level of microscopic counts.

Freezing appeared to reduce the coliform content of melange in much the same way as it did the total count (Table 3). The *E. coli* content, on the other hand, appeared to be more variable and did not show much drop until after 3 months. The reduction in coliforms is, however, much less marked than that reported by Holtman (3).

The studies on the effect of freezing upon the bacterial flora yielded rather inconclusive results (Table 4). Some surprisingly large differences were noted between the various samples in a given lot. There is some indication that freezing may reduce the proportion of *Pseudomonas* species, a definite drop being noted for each of the four samples analysed. This is in agreement with the findings of Lochhead and Jones (12) that organisms in frozen-pack vegetables developing at 4° C. (39.1° F.) were least resistant to freezing. In our studies the number of cultures was too small to warrant the drawing of very definite conclusions.

It was thought that freezing might result in uneven distribution of the organisms within the frozen block. If ice crystals form first at the periphery, the egg solids and accompanying bacteria might be expected to become concentrated toward the center (1, 14). To check on this possibility, borings were made in the center, near the end of the block, and midway between the first two borings. Separate portions were obtained for each boring from the top 2 inches, 2 to 4 inches, and 4 to 6 inches deep. Two blocks, one from Lot D and one from Lot E, were sampled in this manner after 6 months' storage. In addition to plate counts at 30° C., total solids were determined. Contrary to expectations, the highest counts and total solids were found in the top 2 inches, while counts and solids from the center core were lower than those from the end of the block (Table 7). Somewhat similar counts were obtained by Holtman (4).

It will be observed that in both series the level of counts is far above that ordinarily encountered in melange prepared from good quality eggs. Lepper, Bartram and Hillig (11) state, "In no instance did dried eggs show a microscopic count exceeding 10 millions per gram or frozen eggs 5 millions per gram when they were prepared from sound raw material. In all cases where these counts were exceeded, decomposed or rotten eggs had been

incorporated in the product or the eggs had been subjected to conditions after breaking-out which permitted them to sour." In our studies we have good reason to believe that neither of the above-mentioned conditions was responsible for the high level of counts. Only graded eggs, preponderantly Grade A, were used. They had been in storage for from several weeks to 3 months; all were examined for odour and appearance on breaking and any of doubtful quality rejected. Plant sanitation was generally satisfactory in both plants, regular check-ups being made by the resident inspectors and plant laboratories, supplemented by occasional sanitation surveys conducted by the senior author. In no instance were conditions or practices encountered which could conceivably result in counts of the magnitude recorded. Studies reported in another paper (10) suggest that high counts on eggs throughout Canada during the summer of 1944 were attributable to a small percentage of apparently normal eggs which contained very large numbers of bacteria. These bacteria were able to penetrate the intact shell of new-laid eggs and to grow to enormous numbers without causing sufficient change in either odour or appearance to warrant their rejection by the breakers.

SUMMARY

The freezing process brought about a sharp reduction in the numbers of bacteria present in whole egg melange. In two series of tests, involving 44 samples, the average reduction in count was nearly two-thirds. Subsequent storage at 5° F. (−15° C.) for 6 months resulted in little further change in count, although *Escherichia coli* appeared to die off after 3 months.

Plate counts at 37° C. for 2 days were less than a quarter of those at 30° C. for 3 days. Direct microscopic counts were almost always lower than plate counts at 30° C. No explanation for this anomaly has been found.

Freezing and subsequent storage for 6 months appeared to reduce the proportion of *Pseudomonas*, but other genera showed variable results.

There was no indication that the freezing process led to a concentration of bacteria and egg solids in the center of the frozen block.

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TABLE 1.—EFFECT OF FREEZING AND SUBSEQUENT STORAGE ON PLATE COUNTS OF EGG MELANGE. SERIES I

(Counts in thousands per milliliter)

Sample No.	Prepared for freezing	Before freezing		After 48 hours		After 1 month	After 3 months	After 6 months
		A*	B*	A	B	A	A	A
A 1†	May 8	40	7.7	74	12	—	—	—
2		360	58	180	23	—	—	—
3		750	—	320	—	—	—	—
4		340	—	160	—	—	—	—
B 1	June 5	19,000	2,900	4,000	790	3,100	1,000	3,000
2		16,000	3,300	7,300	1,000	6,000	4,400	4,800
3		16,000	3,500	6,600	900	5,900	2,100	4,400
4		15,000	2,500	7,900	1,300	5,500	2,100	5,700
C 1	June 7	7,000	2,100	2,300	530	1,600	2,000	2,300
2		6,900	2,000	1,900	390	1,800	2,500	2,600
3		10,000	2,500	2,500	670	3,300	2,900	2,200
4		12,000	3,800	2,600	500	2,900	3,200	2,900
D 1	July 3	5,000	960	1,100	310	820	290	620
2		6,900	890	1,100	290	840	290	730
3		4,000	960	1,100	430	610	290	880
4		3,400	650	1,100	420	780	520	690
E 1	July 10	10,000	4,100	4,000	740	3,000	3,600	4,500
2		10,000	2,200	4,900	670	4,700	6,900	6,400
3		5,300	1,100	690	150	990	930	1,500
4		16,000	8,000	4,200	710	2,400	4,500	6,400
F 1	Aug. 2	9,900	—	8,600	—	7,900	8,400	8,200
2		9,700	—	6,600	—	6,600	6,300	6,500
3		13,000	—	6,600	—	5,300	3,200	4,400
4		8,100	—	1,400	—	3,500	2,100	1,600

* A = Incubation at 30° C. for 3 days: B = at 37° C. for 2 days.
† This mould was erroneously placed in a warmer room than the other 3, and except for a crust on the surface was still liquid when sampled after 48 hours.

TABLE 2.—EFFECT OF FREEZING AND SUBSEQUENT STORAGE ON DIRECT MICROSCOPIC COUNTS OF EGG MELANGE. SERIES I

(Counts in thousands per milliliter)

Sample No.	Prepared for freezing	Before freezing	After 48 hours	After 1 month	After 3 months	After 6 months
A 1	May 8	<70	<70	—	—	—
2		<70	<70	—	—	—
3		150	<70	—	—	—
4		150	<70	—	—	—
B 1	June 5	7,300	510	880	340	2,300
2		6,300	1,500	1,500	660	2,600
3		6,000	1,700	1,800	660	1,800
4		5,900	1,200	810	880	2,000
C 1	June 7	730	510	880	590	1,400
2		1,700	1,100	590	660	590
3		1,600	1,200	810	590	370
4		2,200	810	590	940	1,500
D 1	July 3	1,800	1,800	340	370	150
2		1,800	1,500	340	290	370
3		1,500	880	290	370	660
4		3,700	2,500	340	730	810
E 1	July 10	2,100	3,100	1,100	5,900	3,400
2		1,500	3,800	2,100	6,100	8,900
3		940	730	590	2,100	1,800
4		3,200	2,000	1,200	3,700	3,300
F 1	Aug. 2	5,100	3,700	5,000	4,500	2,800
2		5,700	3,200	5,800	4,600	5,800
3		5,200	2,200	2,800	1,600	2,100
4		5,800	1,200	2,400	1,800	1,500

TABLE 3.—EFFECT OF FREEZING AND SUBSEQUENT STORAGE ON THE COLIFORM AND *E. coli* CONTENT OF EGG MELANGE. (SERIES I)

(Counts as most probable numbers per 100 ml.)

Sample No.	Before freezing		After 48 hours		After 1 month		After 3 months		After 6 months	
	Coliform	*E coli*	Coliform	*E. coli*	Coliform	*E. coli*	Coliform	*E. coli*	Coliform	*E. coli*
C 1	920	79	240	45	350	20	350	110	240	0
2	240	45	240	45	350	41	240	79	350	18
3	1600	69	350	79	240	45	350	79	350	0
4	920	45	350	79	350	20	240	0	240	0

TABLE 4.—TYPES OF ORGANISMS ISOLATED FROM FRESH AND FROZEN MELANGE. LOT C, SERIES I

Genus	Fresh melange					Melange frozen 6 months				
	C 1	C 2	C 3	C 4	Average for lot	C 1	C 2	C 3	C 4	Average for lot
	%	%	%	%	%	%	%	%	%	%
Proteus	28.2	31.5	33.8	19.0	27.9	12.7	41.0	45.8	41.8	35.1
Flavobacterium	22.5	13.7	36.3	50.0	31.5	17.3	32.0	35.6	33.0	29.5
Pseudomonas	39.5	23.3	16.2	15.5	23.1	20.0	16.7	8.6	12.7	14.6
Achromobacter	5.6	23.3	2.5	10.7	10.4	29.4	7.7	7.1	12.7	14.2
Bacterium	2.8	5.5	10.0	2.4	5.2	7.0	1.3	—	—	2.0
Serratia	1.4	2.7	—	—	1.0	13.4	1.3	2.9	—	4.3
Bacillus	—	—	—	2.4	0.6	1.4	—	—	—	0.3
Sarcina	—	—	1.3	—	0.3	—	—	—	—	—
No. of cultures	71	73	80	84	—	75	78	70	79	—

TABLE 5.—EFFECT OF FREEZING ON COUNTS OF EGG MELANGE. SERIES II. (AUG. 23-25, 1944)

(Counts in thousands per milliliter)

Sample No.	Before freezing		After 44 hrs. freezing		Survival	
	Plate	D.M.	Plate	D.M.	Plate	D.M.
					%	%
1	8,400	3,700	2,300	—	27.4	—
2	7,700	8,600	3,900	11,000	50.6	128.0
3	11,000	6,400	3,800	6,500	34.5	101.3
4	9,500	5,700	5,100	5,900	53.7	103.8
5	10,000	4,800	3,100	2,400	31.0	49.0
6	13,000	7,300	7,600	6,300	58.5	86.3
7	11,000	5,400	3,200	5,200	29.1	96.4
8	16,000	8,500	4,900	4,800	30.6	56.4
9	9,900	7,400	2,700	2,000	27.3	27.0
10	8,700	5,600	2,500	2,300	28.7	41.1
11	12,000	5,600	3,200	3,200	26.7	57.2
12	12,000	5,100	4,200	2,300	35.0	45.1
13	20,000	5,600	6,100	5,600	30.5	100.0
14	19,000	4,800	—	—	—	—
15	13,000	6,400	6,200	2,800	47.7	43.7
16	17,000	11,000	7,400	8,400	43.5	76.4
17	20,000	9,500	10,000	7,300	50.0	76.9
18	16,000	7,800	8,500	5,700	53.1	73.1
19	21,000	9,300	8,700	7,800	41.4	83.9
20	25,000	9,500	7,300	7,300	29.2	76.9
Average					38.34	73.47

TABLE 6.—EFFECT OF FREEZING AND SUBSEQUENT STORAGE ON BACTERIAL CONTENT OF MELANGE. BURRI SLANT TECHNIQUE. DATA FURNISHED BY E. W. NOTON

(Counts in thousands per milliliter)

Carton No.	At start	After 15 days frozen	
		Chip defrosted	Entire contents defrosted
1	100	42	80
	110	—	76
2	310	140	230
	320	—	260
3	330	200	230
	340	—	260
4	280	130	200
	290		160
5	10	12	14
	8	—	12

TABLE 7.—DISTRIBUTION OF BACTERIA AND SOLIDS IN BLOCKS OF FROZEN MELANGE

(Bacteria counts in thousands per milliliter)

—	End of block		Intermediate	Center of block	
	Count	Solids	Count	Count	Solids
		%			%
D 3					
Top 2″	1,400	30.32	1,200	670	31.48
2″–4″	1,100		1,800	760	28.42
4″–6″	1,300		690	630	27.74
E 3					
Top 2″	2,100	28.75	1,100	1,100	29.71
2″–4″	1,200		810	920	27.88
4″–6″	840		840	880	27.14

THE EFFECTS OF A VITAMIN B MIXTURE, OF LEVEL OF PROTEIN, AND OF PROPORTION OF PROTEIN OF ANIMAL ORIGIN IN THE SUPPLEMENTS TO BARLEY AND TO WHEAT IN THE BACON HOG RATION[1]

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Comparative hog feeding tests carried out at this Station have brought to light real differences in feeding value between wheat and barley. Most of the differences have appeared to be consequences of the tendency of wheat to stimulate greater fattening than does barley. This may in part be related to a greater percentage of total digestible nutrients in wheat, but in addition seems to be specifically influenced by the greater content of vitamin B complex of wheat. There is also the possibility that differences in the protein make-up of these two grains affects their nutritive value for hogs. Thus the kind of protein supplement to be used with respect to the proportions of proteins from animal as against plant sources, and the level of the vitamin B complex to be sought by feed selection or specific fortification might differ depending on whether wheat or barley was to be the basal feed.

It was to obtain data on these questions that a feeding trial involving 128 Yorkshire pigs was undertaken in the spring of 1944. The detail of the rations fed and of the plan of the test are shown in Figure 1.

For the first replicate 64 May-born pigs were allotted at random to the several sub-groups, subject to the restriction of equal numbers of males and of females in each treatment group. A second replicate was subsequently fed using January-born pigs.

In each case the pigs were placed on test within a week following weaning at about 8 weeks of age. Throughout the feeding, pigs were confined to individual pens thus permitting individual feed intake records.

The results of the trial were measured in terms of gains of the pigs, their feed consumption under full feeding, shipping weight, and the following carcass characteristics: length of side, depth of shoulder and back fat, leanness of bacon rasher, carcass score and grade.

The length of side was measured in inches from the first rib to the aich bone; leanness of bacon rasher by the square-inch surface area of the "pork-chop" muscle cut between the third and fourth lumbar vertebrae, and also by a planimeter measurement of the surface areas of lean in the full bacon rasher cut as above; carcass score was calculated in accordance with the values used by the Canadian Swine Advanced Registry scheme of carcass evaluations; and carcass grade was set by official graders of the Department of Agriculture.

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[1] Contribution from the Faculty of Agriculture, McGill University, Macdonald College, Que., Canada. Journal Series No. 212.
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FIGURE 1.—DESIGN OF FEEDING TEST

Protein levels, ration composition, and % protein of animal origin				Normal (15.2%) protein level		Reduced (13.4%) protein level	
				5.6 Tankage 5.6 Oilmeal 2.8 Minerals 86.0 Basal feeds	2.8 Tankage 8.4 Oilmeal 2.8 Minerals 86.0 Basal feeds	2.8 Tankage 2.8 Oilmeal 2.8 Minerals 91.6 Basal feeds	1.4 Tankage 4.2 Oilmeal 2.8 Minerals 91.6 Basal feeds
Basal feeds	Vitamin B supplement	Season	Sex	Protein of animal origin 2.8%	Protein of animal origin 1.4%	Protein of animal origin 1.4%	Protein of animal origin 0.7%
No. 2 feed barley	Control	Jan. pigs	♂	2 pigs			
			♀	2 pigs			
		May pigs	♂		2 pigs		
			♀		2 pigs		
	Vitamin B complex*	Jan. pigs	♂			2 pigs	
			♀			2 pigs	
		May pigs	♂				2 pigs
			♀				2 pigs
Durum wheat	Control	Jan. pigs	♂	2 pigs			
			♀	2 pigs			
		May pigs	♂		2 pigs		
			♀		2 pigs		
	Vitamin B complex*	Jan. pigs	♂			2 pigs	
			♀			2 pigs	
		May pigs	♂				2 pigs
			♀				2 pigs

* Mixture added at rate of 0.36 grams per 100 lb. feed intended to supply 3, 5, and 10 milligrams of thiamine, riboflavin, and niacin, respectively, per 100 lb. live weight of pig.

The design permitted an analysis of variance of these items (excepting grade) according to the following scheme:

Source of variation	Degrees of freedom	
All causes		127
Between sub-groups		63
January- vs. May-born pigs	1	
Male vs. female	1	
B complex vs. nil	1	
High vs. low protein	1	
50% vs. 25% animal protein	1	
Barley vs. wheat	1	
Interaction	57	
Within sub-groups (error)		64

The findings of this test may perhaps best be presented according to the separate criteria used. In general only comparisons which show differences of statistical significance or appear to have practical importance will be shown or commented upon.

Seasonal Differences

The effects of season, especially on carcass excellence were of some consequence.

TABLE 1.—MAY- VS. JANUARY-BORN PIGS

Season	Daily gain	Daily feed	Grade A* carcasses	Eye of lean in pork-chop at 3rd lumbar vertebra	Length of carcass, 1st rib to aich bone	Depth of fat: Maximum shoulder fat	Depth of fat: Minimum back fat	Lean in bacon rasher
	lb.	lb.	%	sq. in.	in.	in.	in.	%
May-born	1.19	5.7	55	5.14	29.6	1.70	1.09	42
January-born	1.25	5.1	28	4.42	30.3	1.91	1.35	35

* In Canada a bonus of $3.00 is paid over the base bacon carcass price for each carcass graded A, and $2.00 for grade B.

These seasonal differences include a change in the breeding of the pigs. The January-born and May-born pigs were sired by different boars, and it is evident from other data in our herd that the sire of the January-born pigs caused an increase in the length of the carcasses. This increase in length of side would be expected to be correlated with a leaner carcass and perhaps also with a higher grading. This, however, is not shown in the above data. January-born pigs were fatter, of lower grade, and were considerably faster gaining in spite of lower feed intake.

The May-born pigs were fed for the last month or more of their fattening during cold weather, while the January-born pigs were finished during the summer. Our records show that cold weather results in slowing of rate of gain where pigs are kept in unheated quarters. There is evidence, both experimental[4] and from actual farm practice, that cutting the rate of gain during the latter part of the finishing period results in an increase in carcass leanness. It seems probable that this is the explanation of the seasonal differences in carcass quality noted in this test. (These seasonal effects do not disturb interpretation of the other comparisons, since they were equally distributed to all treatments).

Male vs. Female Pigs

Marked sex differences were found in this trial as shown in Table 2. Males produced fatter carcasses, and as a penalty, fewer grade A's.

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TABLE 2.—EFFECT OF SEX ON CARCASS EXCELLENCE

Season	Daily gain	Daily feed	Grade A* carcasses	Eye of lean in Pork-chop t 3rd ! umbar vertebra	Length of carcass, 1st rib to aich bone	Depth of fat		Lean in bacon rasher
						Maximum shoulder fat	Minimum back fat	
	lb.	lb.	%	sq. in.	in.	in.	in.	%
Males	1.32	5.46	30	4.33	29.7	1.87	1.28	37.7
Females	1.12	5.31	54	5.24	30.2	1.75	1.15	40.0

These findings emphasize the necessity of careful design in tests of this nature to avoid masking or biasing the results of imposed experimental treatments.

The Effect of the Vitamin B Supplement

The effects of supplementary allowances of thiamine, riboflavin, and niacin are summarized in Table 3.

TABLE 3.—EFFECTS OF SUPPLEMENTARY VITAMIN B

Vitamin supplement	Daily gain	Daily feed	Grade A carcasses	Lean of bacon rasher	Maximum shoulder fat
	lb.	lb.	%	%	in.
Barley					
Vitamin B complex	1.26	5.77	40	40.1	1.79
Nil	1.27	5.68	53	39.6	1.75
Wheat					
Vitamin B complex	1.19	5.01	25	36.8	1.92
Nil	1.17	5.08	48	38.9	1.78
Normal protein					
Vitamin B complex	1.27	5.46	34	39.8	1.81
Nil	1.32	5.69	45	38.9	1.78
Reduced protein					
Vitamin B complex	1.18	5.32	28	37.1	1.90
Nil	1.12	5.08	56	39.5	1.77
Normal animal protein					
Vitamin B complex	1.24	5.4	31	38.5	1.91
Nil	1.20	5.3	56	39.8	1.75
Reduced animal protein					
Vitamin B complex	1.21	5.4	34	38.4	1.80
Nil	1.24	5.4	44	38.6	1.78
Vitamin B...all combinations	1.23	5.39	32	38.5	1.85
Nil..........all combinations	1.22	5.38	51	39.2	1.76

The effects of the fortification of the diets with these members of the vitamin B family are interesting and are in accordance with expectation on the basis of theory and of previous findings at this Station.

Extra vitamin B has damaged the average carcass grading, and evidently because of over-finish. Since there were no significant differences in rate of gain or feed intake, it may be concluded that the B complex has stimulated the synthesis of fat from the feed eaten beyond that which would have occurred otherwise.

Furthermore, these effects were more pronounced on the wheat than on the barley rations, and on the reduced than on normal dietary protein levels. The former would be expected in view of the especially high B complex in wheat, while the latter follows from the more fattening character of the lower protein rations.

Normal vs. Reduced Protein

The protein comparison (Table 4) involves both level and kind of protein. The chief significant effect of the high (15%) vs. low (13%) protein levels of the ration was in rate of gain. The pigs on the lower protein level gained 0.15 lb. less per day and required 15 days longer to reach market weight than those on normal protein levels.

TABLE 4.—EFFECT OF PROTEIN LEVEL AND PROPORTION OF ANIMAL PROTEINS IN RATION

Protein level	Normal animal protein		Reduced animal protein		All pigs	
	Grade A carcasses	Daily gain	Grade A carcasses	Daily gain	Grade A carcasses	Daily gain
	%	lbs.	%	lbs.	%	lbs.
Normal protein	47	1.28	35	1.32	42	1.30
Reduced protein	40	1.16	43	1.13	42	1.15
All pigs	43	1.22	39	1.22		

Carcass grades showed some interaction between protein level and the proportion of animal protein. Whether or not this interaction is a real effect cannot be stated from statistical analysis, since "carcass grade" does not lend itself to an analysis of variance.

It will be noted that rate of gain was not associated with any difference in carcass grade. This might be taken as contradictory to the interpretation made relative to differences between May- and January-born pigs earlier in the report. However, examination of the data for the pigs at the 100-pound weight reveals no difference between seasons in the rate of gains up to that time (May-born 1.05 lb.; January-born 1.08 lb.) There was, however, 0.1 lb. more daily gain made by pigs on the high than on the low protein rations. Thus the difference in gains due to protein level extended over the whole feeding period, while the effect of gain on carcass quality has, in other experiments, been found only when rapidly gaining young pigs have been restricted in gain during the *latter part* of their fattening period.

Decreasing the proportion of animal protein from "normal" did not influence rate of gain. Whether or not the slightly higher carcass grade on the higher percentage animal protein is significant cannot be stated since it was not possible to apply statistical analysis to the grading records.

TABLE 5.—COMPARISON OF BARLEY VS. WHEAT AS BASAL FEEDS IN RATIONS SUPPLEMENTED DIFFERENTLY AS TO KIND AND AMOUNT OF PROTEIN AND WITH A VITAMIN B MIXTURE

—	Daily feed eaten	Daily gains		Grade A carcasses	Carcass score	Maximum depth neck fat	Eye of lean of chop at lumbar vertebra
		Observed	Adjusted to equal feed intake				
	lb.	lb.	lb.	%	%	in.	sq. in.
Males							
Barley	5.8	1.35	1.25	31	66	1.82	4.4
Wheat	5.2	1.29	1.34	28	61	1.92	4.2
Females							
Barley	5.7	1.18	1.11	63	74	1.72	5.3
Wheat	4.9	1.07	1.19	45	70	1.78	5.2
Normal protein							
Barley	6.0	1.35	1.20	47	73	1.77	4.9
Wheat	5.2	1.25	1.30	35	68	1.81	4.8
Reduced protein							
Barley	5.5	1.19	1.17	47	67	1.76	4.8
Wheat	4.9	1.11	1.23	37	63	1.88	4.7
50% animal protein							
Barley	5.8	1.28	1.13	44	71	1.81	4.8
Wheat	5.0	1.16	1.32	44	67	1.85	4.8
25% animal protein							
Barley	5.7	1.25	1.15	50	69	1.72	4.8
Wheat	5.1	1.20	1.29	29	65	1.85	4.6
Vitamin B							
Barley	5.8	1.26	1.13	40	69	1.79	4.8
Wheat	5.0	1.19	1.32	25	65	1.92	4.7
No Vitamin B							
Barley	5.7	1.27	1.15	53	70	1.75	4.8
Wheat	5.1	1.17	1.30	48	67	1.78	4.8

Barley vs. Wheat

Comparison of barley vs. wheat under the several different conditions of feeding is summarized in Table 5.

Our results have indicated that larger amounts of barley than of wheat rations are consumed by pigs during the period from weaning to market weight, and in consequence the observed gains are somewhat greater. However, the gain per unit of feed eaten is significantly larger on the wheat rations. It might be thought that some of the barley fed had been wasted, especially by the small pigs, thus giving an apparent but not really greater feed intake, were it not for the larger gains made on the barley.

There can be no doubt that wheat feeding results in lower carcass excellence than is obtained with barley.

Protein level appears to have the same general effect on barley as on wheat rations. Reduction of the animal protein, however, has appar-

ently been more serious with wheat rations than with the barley diets. With the larger allowance of animal protein there was no significant effect related to barley vs. wheat.

The unfavourable results, especially on the wheat rations of vitamin B supplement are clearly evident in carcass grade and depth of shoulder fat.

CONCLUSIONS

1. Season may be an important factor in the excellence of bacon carcasses produced. If pigs are finished in winter in cold pens the rate of gain may be sufficiently reduced to result in a superior carcass.

2. Carcasses from male pigs are by nature fatter than those from females. The tendency for wheat as compared to barley to damage carcass excellence is more pronounced with male than with female pigs.

3. Special supplementation with a mixture of thiamine, riboflavin and niacin tended to aggravate the already greater tendency of wheat than of barley to produce fat carcasses.

4. Reduction of the protein level of the ration from 15% to 13% was reflected in lower feed intake, and slower gains but did not affect carcass excellence regardless of whether wheat or barley was the basal feed.

5. Halving the proportion of the protein from animal origin had no measurable effect either on the progress of the live pigs or on the excellence of the carcasses. Apparently 1.4% of protein of animal origin in the ration—equivalent to only 9% of the protein of a ration carrying 15% protein—is as satisfactory as is double this amount.

ACKNOWLEDGMENT

We are indebted to the Canadian Co-operative Wheat Producers for their continued interest and financial support of these studies on the feeding values of the cereal grains for swine, and to Canada Packers for the use of abattoir facilities and for absorbing losses incidental to cutting the carcasses.

SUPPLEMENT TO
PAMPHLET No. 22

ISSUED FEBRUARY, 1946

DOMINION OF CANADA—DEPARTMENT OF AGRICULTURE
EXPERIMENTAL FARMS SERVICE

CORN HYBRIDS RECOMMENDED FOR PRODUCTION IN ONTARIO—1946

VERY EARLY

Canbred	150
Canada	240
Wisconsin	240
Canbred	250
Canada	255
Wisconsin	255
Canada	275
Wisconsin	275
Canada	279
Wisconsin	279
Wisconsin	335
Canada	355
Wisconsin	355

MEDIUM

Funks	G550W
Harvic	300
DeKalb	240
Pioneer	353A
Canada	606
Wisconsin	606
Canada	645
Wisconsin	645
Canada	625
Wisconsin	625
Harvic	333
Pfister	274
Pioneer	373
Funks	G31
Jacques	1157
Hoosiercrost	F138

EARLY

Wisconsin	416
Funks	G184
Harvic	222
DeKalb	65
Funks	G176
Pioneer	355
Wisconsin	531
Canada	531
Iowa (White)	3215

LATE

Funks	G15
Pioneer	322
DeKalb	404A
Indiana	210B
Canada	696
Wisconsin	696
Wisconsin	692
Pfister	4897
Pride	D66
Top Crop	115
DeKalb	458
Pioneer	340
Iowa	939
Funks	G29
Illinois	972
Indiana	610

NOTE—Hybrids designated "Canada" have the same parentage as "Wisconsin" hybrids of the same numbers. They are produced in Canada from breeding stocks now maintained in Canada but which originated in Wisconsin.

The hybrids are grouped according to maturity, with the earlier hybrids at the top in each group. Growers will appreciate that maturity may vary somewhat under different soil and climatic conditions.

The hybrids have been selected to meet a wide range of conditions. All of them have demonstrated high yielding ability and possess strong stalks and large root systems. They have shown considerable resistance to lodging, and to damage resulting from corn borer attacks. All have shown themselves to be superior to the commonly-grown varieties of similar maturity.

Note—This is a continuation of War Series Pamphlet No. 22. Data compiled by the Ontario Corn Committee.

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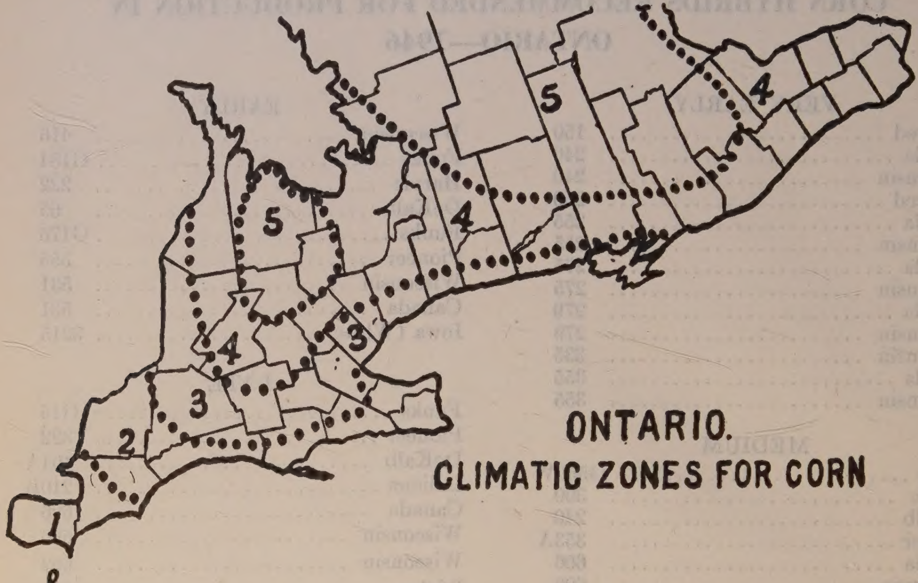
Zones of Production

The zones outlined on the map are based upon the average number of frost-free days and the total number of heat units (1) registered during the growing season. In drawing the zone boundaries the fact is fully recognized that there are many local areas within each zone, in which the soil and climate differ somewhat from the average of the zone. Nevertheless, the zone boundaries will provide a useful guide for the inexperienced grower who wishes to choose a corn hybrid suitable for production in his particular area.



(1) Meteorological data supplied by the Ontario Research Foundation.

Recommendations Based on Zones

ZONE 1
- Fodder .... Late group adapted to whole area.
- Grain .... Medium group adapted to whole area. Late group preferable in many localities.

ZONE 2
- Fodder .... Late and medium groups adapted to whole area.
- Grain .... Early group adapted to whole area. Medium group preferable in many localities.

ZONE 3
- Fodder .... Medium group adapted to whole area. Late group preferable in many localities.
- Grain .... Very early group adapted to whole area. Early group preferable in many localities.

ZONE 4
- Fodder .... Medium or early group adapted to whole area.
- Grain .... Very early or early group adapted to many localities.

ZONE 5
- Fodder .... Early or very early group adapted to many localities.
- Grain .... Very early or early group adapted to favoured localities.

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